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Project Thesis

Clinical use of targeted alpha particle therapy, a literature review.

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1.0 Abstract

Objectives: Targeted Alpha Therapy (TAT) is a cancer treatment technique involving the use of an alpha-emitter and a vector conjugate, combining the short-range high toxicity properties of an alpha-emitting radionucleotide, and the ability to directly target cancer tissue of a vector (for example a monoclonal antibody). TAT is a form of personalized treatment and its design is aimed to be especially effective against disseminated cancer, making it one of few options against late-stage cancer. Although the technique has been proposed since the late 1990s, little progress have been made until recently, with a number of clinical trials on going.
The main aim of this thesis is to provide a literature review of current published studies of clinical trials utilizing Targeted Alpha Therapy. The second aim is to provide an overview on safety and efficacy results across the aforementioned articles.

Method: A literature search was conducted via the Ovid search engine in the following databases: *Embase Classic+Embase (1947 to 2023 January 06)* and *OVID MEDLINE(R) ALL (1947 to January 06, 2023)* on the 9th of January 2023. The preliminary results of the search yielded 161 items. The number of articles that met the inclusion criteria after three screening processes (Figure 2) was 13. These 13 articles were included in this literature review.

Results: Thirteen articles describing clinical use of targeted alpha therapy were found, of which two were pilot studies, seven were phase 1 clinical trials, and four were retrospective analysis of case series (Table 1). The studies included between 5 to 28 patients (Table 2) and TAT was given to patients with five different types of cancer (one type per article). The types of cancer covered in the 13 articles are: metastatic castration-resistant prostate cancer (mCRPC), glia tumors, neuroendocrine tumors (NETs), leukemia, malignant melanoma and carcinoma in situ of the bladder (Table 2). Safety and efficacy results are summarized in Table 4, and the main conclusions from the original articles are summarized in Table 5.

Conclusions: The safety profiles among the 13 articles show some variation, most likely due to the heterogeneity across the studies (patient populations, types of cancer treated, TAT products and study protocols used). Five of the studies reported no treatment emerged adverse effects (TEAEs as by common terminology criteria for adverse events (CTCAE)). Whereas the remaining eight studies report some TEAEs, most of them being mild (Grade 1-2). Few severe adverse events where reported, most of which were considered not related to treatment with TAT.
Generally, regarding safety profiles, TAT seems to have little toxicity with manageable side effects (Table 4). When it comes to efficacy, the results also show variability, with some patients having little effect of treatment while others showing remarkable results (Table 4; Figure 3 and 4). This not being too discouraging since TAT is a form of personalized treatment that is under development. Improvement in efficacy outcomes is likely after further investigation and updated treatment protocols.
To summarize, TAT seems to have promising preliminary efficacy results and manageable toxicity, something that should warrant further investigation.

2.0 Abbreviations

- TAT – Targeted Alpha Therapy
- CTCAE (Common Terminology Criteria for Adverse Events)
    - AEs – Adverse Events
    - TEAEs – Treatment Emerged Adverse Events
      (AEs/TEAEs are classified as per CTCAE into five severity grades:
      1 = mild, 2 = moderate, 3 = severe, 4 = life threatening, and 5 = death).
- mCRPC – Metastatic Castration-Resistant Prostate Cancer
- NETs – Neuroendocrine Tumors
- PSMA – Prostate-specific membrane antigen
- SMLs – Small Molecule Ligands
- mAb (mAbs) – Monoclonal Antibody/(Antibodies)
- RECIST 1.1 – Response Evaluation Criteria in Solid Tumors version 1.1
    - ORR – Overall Response Rate
    - CR – Complete Remission
    - PFS – Progression Free Survival
    - OS – Overall Survival
    - PR – Partial Remission
    - SD – Stable Disease
- FDA – U.S. Food and Drug Administration
- EMA – European Medicines Agency
- IV – Intravenous Administration
- SPECT – Single Photon Emission Tomography
- CT – Computed Tomography
- MR – Magnetic Resonance imaging

3.0 Introduction

3.1 Introduction to Targeted Alpha Therapy

Targeted Radionuclide Therapy and Targeted Radiotherapy includes both alpha- and beta labelled tracers as radiation source. Targeted Alpha-particle Therapy (TAT), also known as Targeted Alpha-emitter Therapy and Targeted Alpha Therapy is a technique that is in rapid development in oncology. It can theoretically be used against most types of cancer (1). Treatment of disseminated cancer is one of the main challenges in oncology, and TAT offers a new and potentially revolutionizing gateway to improving treatment outcomes for late-stage cancer patients.

TAT is a form of personalized treatment. Patients are tested for the presence of biologic targets on their tumors (e.g. cell surface or stromal components) (2). Upon confirmation that a specific vector (for example a monoclonal antibody) can target the patients tumor/s, an alpha-emitting nucleotide is conjugated to the vector. The effectiveness of TAT can be explained by the properties of alpha-emitting nucleotides (alpha-emitters) that release enormous amounts of energy over a very short distance. Combining the effectiveness of high toxicity with a short range (of an alpha-emitter), with the specific targeting abilities of a vector. This way one can specifically target cancer cells and treat hundreds of metastases simultaneously, as TAT delivers potent toxicity (high radiation dose) to target lesions, with minimal damage to the surrounding healthy tissue (3).

- This chapter provides only a brief introduction to TAT. A more comprehensive discussion of the technique is presented in Chapter 3.3.3.

3.2 Thesis goal and results summary

Due to their cytotoxic abilities, alpha-emitters have been suggested for clinical use since their discovery by Rutherford in 1898 (1). However, the development of adequate techniques that can be applied clinically has been somewhat slow. The first clinical study with a targeted alpha-emitter dates back to 1999 (4), but the development of a full product took time, probably due to the high cost and low availability of alpha-emitters as well as limited options for vectors and chelators. The first and only approved TAT radiopharmaceutical is Xofigo® (approved for mCRPC in 2013 (5)). Recent breakthroughs in generating alpha-emitters, chelating techniques and vector technology have made TAT increasingly attractive. Several studies have demonstrated promising results, and many more are underway.

The goal of this thesis is to present a Literature Review on all published clinical trial studies involving a form of TAT. We chose to exclude articles describing data from the approved product Xofigo® ($^{223}RaCl_2$), and instead focused on all other alpha-emitter products studied.

The literature search (conducted on 09.01.2023) yielded thirteen eligible articles describing the use TAT against several different types of cancer (metastatic castration-resistant prostate cancer (mCRPC), glioma, neuroendocrine tumors (NETs), leukemia, malignant

melanoma and carcinoma in situ of the bladder). The articles included in the current thesis were either in early stages of clinical trials (pilot studies, phase 1 studies) or retrospective analysis of case series: The focus among the thirteen studies was primarily on safety profiles, with most studies having a dose expansion protocol. Although treatment efficacy was not the main focus, preliminary results indicate promising treatment outcomes, especially among patients receiving higher doses. These findings suggest that a better understanding of treatment doses and improved protocols could lead to even better patient outcomes.

Generally, regarding patient safety profiles across the thirteen studies, we mostly have mild and manageable treatment emerged adverse events (TEAEs / side effects / toxicity), with very few serious TEAEs reported, most of which were considered not related to TAT. The original authors seem to conclude that the TAT is most often well tolerated and the TEAEs are manageable (Table 5). Note also that all the original authors seem to be in agreement that TAT is a promising method and warrants further investigation and research.

When it comes to efficacy results, there was some variation among patients. While some patients seemed to have minor effect from treatment, some patients showed remarkable treatment results. The fact that not all patients had treatment effect is truly not discouraging, considering that TAT is a personalized treatment method, and not all patients are expected to qualify for treatment. Moreover, treatment doses and protocols are still not fully developed yet. However, what is extremely interesting are the results on some patients where the treatment effects are remarkable. An example is the MAD4 cohort patients from the Delpassand study (6), with metastatic NETs that show exceptional tumor regression after four cycles of 212Pb DOTAMTATE. (see Figure 3 and 4)

The completed clinical trial studies with TAT, serve as a proof of concept that TAT has manageable toxicity and can be effective in treating different types of cancer. TAT is most definitely promising, and early clinical results indicate that it could potentially revolutionize treatment outcomes for persons with late-stage cancer, who often have few to no options regarding treatment.

3.3 Theory and background

3.3.1 Radiation and radiation types

Radiation is energy coming from a source and traveling through space or a medium. The energy can be both in the form electromagnetic waves (rays) or particles. We often speak of ionizing radiation and non-ionizing radiation. Non-ionizing radiation has less energy and unlike ionizing radiation, non-ionizing radiation does not remove electrons from atoms or molecules (7). Examples of non-iodizing radiation are radio waves, visible light, microwaves, infrared light etc.

Clinically, it is interesting to look at ionizing radiation as it can affect atoms, molecules in the human body. Ionizing radiation comes from radioactive atoms. Which are unstable in the meaning that they have an unbalanced number of protons and neutrons in their nucleus.

Atoms seek to be stable and therefore they expel energy from their nucleus in the form of particles or ray, and this is called radiation (8).

The ionizing radiation coming from radioactive atoms is typically created by radioactive decay. A radioactive atom will attempt to reach stability by ejecting nucleons and other particles, or by releasing energy in other forms, often as rays. The following are some of the main types of ionizing radiation (note there are also other types of radiation beside the below mentioned, both as particles and rays):

- α-radiation/α-particles, that come from α-decay are basically helium ions made up of two neutrons and two protons with a charge of +2. The range of the α-particles is very short in matter. The α-particles can be stopped by a piece of paper, a few centimeters of air, or gloves.
- β-radiation is also particle radiation, but with much smaller β^+ or β^- particles. β^--particles are electrons emitted from the β^--decay where a neutron is converted to a proton in the nucleus. β^+-particles are positrons and are emitted through β^+-decay when a proton is converted to a neutron and a positron. (9) (Positrons are the antimatter of electrons, meaning they have the same mass but a positive charge.) Beta particles can be blocked effectively with a few inches of plastic, or even a layer of clothing.
- γ-radiation or γ-rays are high-energy electromagnetic waves that expel the extra energy that arises from isomeric transition from an upper energy state of a nucleus to a lower energy state. The energy of the γ-ray emitted is the difference between the two isomeric states. γ-rays can travel through matter. It takes several inches of lead or other dense substance to block gamma rays. However, they possess less energy then α- or β-particles (9).
- X-rays are also electromagnetic waves that expel excess energy. The main difference between X-rays and γ-rays is that X-rays are emitted from processes outside the nucleus, while γ-rays originate from inside the nucleus. It takes only a few millimeters of lead to stop x-rays (9).

Figure 6 summarizes and illustrates the abovementioned types of radiation.

Heavy atoms, that are unstable, decay until they reach a steady state. A mother nucleotide decays often to multiple daughter nucleotides in a chain reaction expelling different types of radiation and producing a new daughter nucleotide (each with its own half-life) on each step of the reaction. Figure 1 illustrates this by depicting the decay scheme of several alpha-emitters (Figure 1, (10)). Here can one se both that we have creation of a number of daughter nucleotides, and that several types of radiation are expelled.

3.3.2 Cytotoxicity of radiation

As radiation is energy, it interacts with matter as it passes through it until its energy is depleted. Ionizing radiation ionizes or excites the absorbers´ (the matter it goes through) atoms. Radiation cytotoxicity in living organisms lies primarily in its ability to introduce irreparable damage in double-stranded DNA.

Due to their ability to destroy tissue, α- and β- radiation particles were quickly applied to therapeutic applications such as radiation cancer therapy, since their discovery in the late 1898 by Rutherford, where β- radiation particles have been the preferred medium. (1)

Cytotoxic effect depends on the type of radiation, the energy it carries and the amount of energy it deposits as it goes through matter. To define this, we use the term "Linear Energy Transfer (LET)». LET is the amount of energy deposited per unit length of the path by radiation. (LET = SI × W; unit: keV/μm).
Here lies the distinction between α- and β- radiation. The α-decay pathway releases enormous amounts of energy over a very short distance. Typically, the range of α-particles in tissue is 50–100 μm, and they have high linear energy transfer (LET) with a mean energy deposition of 100 keV/μm (LET). Which is much higher than that of β-particles that have a mean energy deposition of 0.25 keV/μm (LET), and a range of 3500-4000 μm in biological tissue. (9).
The following example illustrates this difference: The mean LET value for the β-particle-emitting ^{90}Y (Yttrium-90) is 0.2 keV/μm whereas that of α-particle emitting ^{211}At (Astatine-211) is 97 keV/μm. Furthermore, the mean range in tissue of ^{211}At (α-particle) and ^{90}Y (β-particle) are 70 μm and 3960 μm, respectively (3). Cell survival studies have shown that a single (or few) α-particle track, originating from the cell's surface and traversing the nucleus, is capable of resulting in cell death (11). This is in contrast to the thousands to tens of thousands of β-particles required. Therefore, the cytotoxicity induced by alpha particles is far more effective and selective.

3.3.3 Targeted Alpha Therapy (TAT)

Targeted Alpha-particle Therapy (TAT) is a technique that is in rapid development in oncology. It can theoretically be used against most types of cancer. Regardless of if there is one primary tumor or numerous disseminated metastases (1).

TAT has been proposed since the 1990s (12). The nature and characteristics of alpha-particle emitters (short-range, high-energy release) makes them an attractive option for selective targeting of tumors/metastases. The ability to specifically target numerous tumors simultaneously, and fact that it can be combined with other therapeutic approaches (biological, chemo, surgical or other radiation treatments) can make TAT the perfect "magic bullet" (concept first introduced by Paul Ehrlich in 1907 (13)) against disseminated cancer.

TAT peruses a personalized approach. Patients are tested for the presence of biologic targets on their tumors (in cell surface or stromal components) (2). Upon confirmation that a specific vector (for example a monoclonal antibody, or ligand for surface receptors) can target the tumors, an alpha-emitting nucleotide is conjugated to the vector.

The effectiveness of TAT can be explained by the properties of alpha particles that release enormous amounts of energy over a very short distance (9). Targeted alpha-emitter therapy combines the effectivity of high toxicity with a short range (of an alpha-emitter), with the targeting abilities of a vector. This makes it possible to induce selective toxicity with a high radiation dose, almost only on cancer tissue, with limited exposure on the surrounding healthy area.

There are already a number β-emitters approved for use as targeted radiotherapy (for example: ^{131}I for Graves' disease (14), ^{89}Sr for bone metastases, (15); 177Lu-PSMA- 617 against mCRPC, (16). Although β-emitters are more widely used, the properties of α-emitting nucleotides should make for better options as they offer a substantially increased specificity, and by such reducing side effects and improving treatment outcomes.

TAT was first proposed in the 1990s (12), and the interest in TAT has been growing. The first approved product utilizing an α-emitter as therapy came in 2013 and was developed in Norway by Roy Larsen and Øyvind Bruland. After the publication of the ALSYMPCA trial (5), Xofigo® (^{223}Ra-Cl_2) was approved by the Food and Drug Administration (FDA) and later by the European Medicines Agency (EMA) as palliative treatment for castration-resistant prostate cancer patients with symptomatic bone metastases (mCRPC). To this day this remains the only approved α-emitting therapy agent available. ^{223}Ra-Cl_2 is technically not a targeting agent, but a calcium mimic and α-particle emitter that targets areas of increased bone turnover (see chapter 3.6.7).

Nevertheless, the interest in TAT has been substantial for many years, and a number of α-emitting nucleotides such as ^{211}At, ^{212}Bi, ^{213}Bi, ^{255}Fm, ^{212}Pb, ^{225}Ac, ^{149}Tb, ^{223}Ra (17) have been suggested for clinical use. The relatively short half-life of several alpha-emitters, (for example: 46 min for ^{213}Bi, 10.6 hours for ^{212}Pb (1)) has halted the development of TAT products by limiting its use in many locations where the production cannot happen near the clinic. This issue, together with the relatively high cost and low supply of alpha-emitting radionucleotides, has obstructed the development of TAT as a practice.

Recent breakthroughs in radionucleotide production technique, together with the advances in mAB (monoclonal antibody) technology, chemical labeling techniques, dosimetry technology, as well as better understanding of carcinogenesis and cancer biology, has led to significant progress in the development of new clinical options for targeted alpha-particle therapy, with several clinical trials ongoing.

3.3.4 The uncertainty daughter nucleotides.

As described earlier radioactive nucleotides decay through a chain reaction until they reach a steady state. Through this decay process, several daughter nucleotides (decay products) are crated and exist for a certain amount of time. The number as well as the properties of the daughter nucleotides can vary. One can predict to a certain degree how they will act in vivo, but still much is unknown as to what actually happens.

Regarding TAT products, this has to be taken in consideration. When synthetizing a TAT compound one conjugates a radioactive nucleotide to a targeting vector, via a chelator. This is done via different chemical bonds that work in accordance to the chemical and physical properties of the two components they connect (the radionucleotide and the vector). However, as described earlier, the state of a radioactive nucleotide is somewhat dynamic, with new daughter nucleotides taking the place of the old ones, via the decay process (see Figure 1).

Questions that would naturally arise are: What happens with the conjugation and the chemical bonds of the TAT compound, when one of its components suddenly changes? What happens with the daughter nucleotides?; Do they remain on target?; Do they disassociate from their vector in vivo?; Do they diffuse to other organs?
The answers are partly still unknown, and there is an unmet need for further studies.

For example, we know that in ^{223}Ra disintegration scheme, at some point one of the decay products is ^{219}Rn (Figure1; (10)). Radon-219 (^{219}Rn) is a gas that can diffuse through cells. Short lived as it might be, it still opens a possibility for the radio-nucleotide to disassociate from its target (1).
As a safety measure against this particular issue, there are often placed Rn sensors in the rooms of patients receiving $^{223}Ra\text{-}Cl_2$ (Radium-223-dichloride).

3.3.5 The importance of vectors in TAT

When designing a TAT product, one has not only to evaluate and choose the right radionucleotide (alpha-emitter) but also the right vector. Compounds that can act as vectors can be divided into two main groups: (a) monoclonal antibodies (mAbs), that are relatively big proteins/fragments; and (b) ligands (also called SML, small molecular ligands), that are often smaller molecules that bind to specific biological receptors on target cells. (Other substances have also been tried as vectors, though mainly in non-human trials. Examples: colloids, microspheres, liposomes, polymer particles (MDPPs), other molecules (α-methyltyrosine, astatodeoxyuridine etc.) (12).) The vector (together with the chelating agent) directly affects the pharmacodynamics as well as pharmacokinetics of the radionucleotide-vector-conjugate.

To demonstrate the clinical significance of the vector, we can look at reports of beta-emitters used as treatments for mCRPC (metastatic castration-resistant prostate cancer). Prostate-specific membrane antigen (PSMA) is expressed by most prostate cancers and can be targeted by both antibodies (mAb, for example J591) and small molecule ligands (SMLs / for example PSMA-617), each with distinct binding sites, kinetics, and distributions. mAbs are larger, with longer circulating times in the blood, resulting in greater exposure to bone marrow, but lesser access to PSMA expression on luminal tissue (e.g. salivary glands, small bowel, and kidney). In contrast, SMLs are rapidly excreted via kidneys and readily diffuse to all PSMA-expressing sites (18,19).

What this translates to, when adding the radio-emitting nucleotide in the mix, is a difference in side effects and toxicity profiles. Niaz reports this phenomenon in his study, where safety and efficacy of the betta emitting ^{177}Lu (a beta-emitter) is compared between two groups of men (161 receiving 177Lu-J591 and 50 receiving 177Lu-PSMA-617) (18). In the treatment emerged adverse events (TEAEs) report, hematologic TEAEs were more common with 177Lu-J591 (mAb vector): neutropenia (77.9% - mAb vs. 4% - ligand); anemia (77.1% - mAb vs. 16% - ligand), thrombocytopenia (90.1% - mAb vs. 20% - ligand). Non-hematologic TEAEs were more common with 177Lu-PSMA (ligand vector): xerostomia in (58% - ligand vs. 0% - mAb) and nausea in (42% - ligand vs. 14.5% - mAb) (18).

Even though the radionuclide used in the prior example is a beta-emitter and not alpha-emitter, it theoretically applies to both. Therefore, that example perfectly illustrates how the vector directly affects the safety/toxicity profiles of the radionuclide therapy products. By selecting the appropriate vector one can elegantly design a TAT product with an even higher degree of specificity. This making the vector an important aspect in designing TAT-products.

3.3.6 Doses of radioactivity and measuring treatment outcomes.

The unit of radioactivity is Curie (Ci). The SI (The System Internationale) unit for radioactivity is the Becquerel (Bq), which is defined as 1 dps (disintegration per second). (9)

$$1\ \text{Ci (curie)} = 3.7 \times 10^{10}\ \text{dps (disintegrations per second)}$$
$$1\ \mu\text{Ci (microcurie)} = 3.7 \times 10^{4}\ \text{dps}$$
$$1\ \mu\text{Ci} = 3.7 \times 10^{4}\ \text{Bq} = 37\ \text{kBq} = 0.037\ \text{MBq}$$
$$1\ \text{MBq (Megabecquerel)} = 27.027\ \mu\text{Ci}$$

Many studies where small doses of radiation are given to mammalian/human subjects the preferred unit is microcurie (μCi) or megabecquerel (MBq). In the studies we discuss in this review, all the units have been converted to MBq.

Keep in mind that when using these units (MBq and μCi) we describe the total estimated dose of radiation delivered to a subject. We are not discussing the radiation released from individual nucleotides, but rather the cumulative dose of radiation a patient is expected to receive during a cycle of radiation therapy. While this may be less precise in the case of TAT, which is targeting specific tissues, it remains a valuable tool for comparing dosages across treatment cycles. One would have to use microdosimetry methods to further obtain data on the doses of radiation specific tissues receive.

In the articles discussed in this thesis other methods have been used to obtain efficacy data, instead of measuring radiation delivered to target through microdosimetry. Patient outcomes were measured my taking MR imaging or positron emission tomography/computed tomography (PET/CT) with different tracers.

By analyzing the pre- and post-treatment images, one can gain a comprehensive understanding of the extent to which a treatment has affected target lesions.

3.6.7 $^{223}Ra\text{-}Cl_2$ a short review

As per the reasons presented in Chapter 4.2, we have chosen not to discuss $^{223}Ra\text{-}Cl_2$ in this literature review. However, we recognize that Xofigo® ($^{223}Ra\text{-}Cl_2$) is the first and only approved product utilizing alpha-emitter therapy. Therefore, we present here a brief introduction to Xofigo®. As previously mentioned this product became FDA and EMA approved in 2013 after the ALSYMPCA trial (5), a phase III, randomized, double-blind, placebo-controlled study, with 921 patients enrolled. Results confirmed the radium-223 survival benefit (median, 14.9 months vs. 11.3 months; hazard ratio, 0.70; 95% CI, 0.58 to 0.83; P<0.001)(5).

223Ra is used as treatment against mCRPC (metastatic castration-resistant prostate cancer). Deaths from prostate cancer are often due to bone disease and its complications (5,20).

Radium 223 acts a bone-seeking calcium mimetic, and is bound into newly formed bone stroma, especially within the microenvironment of osteoblastic or sclerotic metastases. Thus, Radium-223 dichloride works as an alpha-emitter that selectively binds to areas of increased bone turnover in bone metastases and emits high-energy alpha particles of short range (21),(22).

4.0 Method

4.1 Literature search strategy

A literature search was conducted on the 9th of January 2023 via the Ovid search engine in the following databases:

Embase Classic+Embase (1947 to 2023 January 06)
OVID MEDLINE(R) ALL (1947 to January 06, 2023)

The goal was to find articles describing clinical trials of all stages that used TAT on human subjects. The search was conducted with the following algorithm on both databases simultaneously:

Embase Classic+Embase <1947 to 2023 January 06>
Ovid MEDLINE(R) ALL <1946 to January 06, 2023>

1	alpha particles/ or alpha radiation/ or (alpha particle* or alpha radiation*).mp.	13557
2	(target* and (alpha emitt* or alpha particle*)).mp.	3645
3	(radioimmunotherap* or radiotherap* or therap* or treatment*).mp.	24173676
4	1 or 2	14426
5	3 and 4	5170
6	limit 5 to clinical trial	167
7	6 use emczd	153
8	6 use medall	14
9	7 or 8	167
10	remove duplicates from 9	161

4.2 Article inclusion strategy

Figure 2 illustrates the article inclusion strategy.

Preliminary results yielded 161 potentially eligible studies, that were transferred to the reference management (library) software Zotero.

Inclusion criteria was determined to be:

- Articles based on data gathered from treatment of human subjects (in vivo treatment / Clinical Data).
- Original articles (not reviews or literature studies).
- We choose to include all phase I, II and III studies, pilot studies, as well as retrospective analysis of reports of case series of patients treated with any alpha-emitting agent.

Exclusion criteria was decided to be the following:

- Language (not English).
- Not in vivo data.

➔ Screening no 1: At this point we read the title and abstract of 161 articles. The articles that fulfilled the criteria were grouped in a new sub-library containing 73 articles describing clinical use of alpha-emitters.

➔ Screening no 2: At this point we choose to introduce three additional exclusion criteria:

- Articles describing boron neutron capture therapy were excluded, as this type of therapy needs an external irradiation with low-energy thermal neutrons to yield alpha particles. (4 studies)
- Articles describing the use of ^{223}Ra-dichloride against metastatic castration-resistant prostate cancer (bone metastases) were excluded (43 studies).
  This exclusion criteria were decided upon for two reasons:
  - Firstly, this is already an FDA and EMA approved therapy for bone metastases.
  - Secondly because ^{223}Ra-dichloride is not a "genuine" TAT method. ^{223}Ra-dichloride mimics calcium and is therefore selectively deposited in the skeletal system. It forms complexes with the bone mineral hydroxyapatite and accumulates in bone tissue, especially in areas of high turnover such as skeletal metastases. However, there is no vector used here, the targeting abilities of this product lie in its own chemical properties.
- Articles describing the use of ^{223}Ra-dichloride for other bone metastases (not only prostate cancer) (4 studies).

We reviewed the title and abstract of 73 studies selected in the first screening. Out of these, we excluded some articles based on the new exclusion criteria and compiled a new sub-library of 22 articles that met our inclusion criteria.

➔ Screening no 3: The 22 articles were thoroughly read with data summarized in an excel sheet. Of the 22 articles 9 articles were excluded for the following reasons:

- Article not describing use of an alpha-emitter (n=3).
- Ongoing clinical study without published results (n=3).
- Article not describing clinical data (n=2).
- Article using same pool of patients as another article already used (n=1).

This brought the number of eligible articles to 13. The number of articles we discuss in this literature review are 13 (Figure 2).

5.0 Results

The results from the 13 articles have been summarized in the following tables. The order of articles across all the tables is the same. The data was originally logged on one big excel sheet, and then later divided in 5 tables to make it easier to cross refence the data.

Results are summarized on **Tables 1-5** (In brackets: the column headers of the tables)

- **Table 1** gives general information on the 13 articles included in the discussion (Article Reference Nr; Article Title; All Authors; Type of Study; Article Size; Publication Medium; City/Country of Study; Publication Year).
- **Table 2** summarizes the patient group of each study, and depicts the TAT used (Article Reference Nr; Nr of Patients (in final data review); Alpha-emitter (+ Chelator) + Vector (mAb); Alpha-emitter (+ Chelator) + Vector (ligand); Type of Cancer (on patients enrolled in study)). Note that the TAT-product column is divided in two to make it easier to distinguish whether the vector used is a ligand or mAb.
- **Table 3** focuses on (estimated) doses of radioactivity the Alpha-emitters administered delivered to patients (Article Reference Nr; Alpha-emitter type + vector; Cumulative Dose Range (Mbq/kg); Dose range for one cycle (MBq/kg); Absolute Cumulative Dose Range (Mbq); Dose range for one injection (Mbq/Injection); TAT administration method).
- **Table 4** focuses on the reported Safety and Efficacy data reported form each article (Article Reference Nr; Safety (TEAE: Treatment-Emerged Adverse Events; considered by author related to TAT treatment); Efficacy (objective responses to treatment)).
- **Table 5** focuses on the authors main conclusion on each article (Article Reference Nr; Conclusion).

- The order of the articles on all tables is such that articles treating patients with the same type of cancer are neighboring.
- In all tables the rows have either blue or orange background color.
    - The **blue color** indicates that the administration method of the TAT-product is **intravenous (IV)**, in that particular study.
    - The **orange color** indicates that the administration method of the TAT-product is **intralesional** (intralesional, intratumoral, intravesical) in that particular study.

5.1 The article pool

Thirteen articles were found eligible and included in this literature review (Table 1; (6,23–34)). Of the 13 articles, six were only abstracts (approved for oral sessions/presentations or poster sessions/presentations in international/national conferences and/or journals) (Table 1, column 5). The six articles containing only abstracts were found in: Journal of Clinical Oncology (a peer reviewed medical journal of the American Society of Clinical Oncology Journal / 2 articles: (28,34)), European Journal of Nuclear Medicine and Molecular Imaging (3 articles: (24,25,32)), and Nuklearmedizin, (an annual conference of the German Society for Nuclear Medicine / 1 article: (29)). We chose to include the data from the aforementioned articles in the discussion for two reasons: (a) they are published in peer

reviewed and established journals; and (b) the data obtained and study method was found relevant to this literature review.

Of the 13 studies, eight had an intravenous (IV) administration of a TAT conjugate (blue rows, all Tables), while five used an intralesional approach (intertumoral and intravesical (one article)) (orange rows, all Tables). The intralesional administration-based studies were selected to be included in this review as they also included a systemic focus on TEAE (treatment-emerged adverse events).

Of the 13 studies included, two were pilot studies; seven were phase 1 open label non-randomized studies (six of which had a dose expansion protocol); and four were retrospective analysis of case series (two of which had a dose expansion protocol). (Table 1, column 4)

5.2 The patient pool

In the selected 13 studies there were a total of 202 patients enrolled (range 5 – 28 patients/study). One-hundred and fourth-nine patients received a TAT-product by IV injection/s (in a various number of cycles) (Table 2, column 2 and 5). Fifty-three patients received local injection/s with TAT (of which 41 received intralesional/-tumoral administration and 12 received intravesical administration) (Table 2, column 2 and 5).

All the included patients had cancers. Two articles had patients with NETs (neuroendocrine tumors / total of 31 patients); three studies had patients with mCRPC (castration-resistant prostate cancer with metastases / total of 60 patients); two studies had patients with leukemia (total of 36 patients); two studies had patients with malignant melanoma (total of 38 patients); one study had patients with carcinoma in situ of the bladder / total of 12 patients); and three studies had patients with gliomas (total of 25 patients). (Table 2, column 2 and 6).

5.3 TAT and alpha-emitters used.

In the 13 studies we are to discuss the following alpha-particle emitters have been utilized: ^{212}Pb, ^{225}Ac and ^{213}Bi (Table 2, column 3 and 4). Each study had designed a specific combination of: (a) one alpha-emitter, (b) one chelator, and (c) one targeting vector. In 7 of the 13 studies the vector used was a ligands (-TATE, PSMA, Substance P), while in the remaining 6 the vector used a monoclonal antibody (mAb). (Table 2, column 3 and 4),
On only 8 of the 13 studies was the type of chelator (used to bind the alpha-emitter component to the vector) specified, while in the remaining 5 the chelator was not specified. (Table 2, column 3 and 4).

5.4 Doses of radiation received by the enrolled patients.

Table 3 present the administered doses. All the units used across the different studies (KBq, GBq, Ci, µCi) have been converted to the unit of MBq (mega becquerel), to make the comparison among the articles easier.

Given that the 13 studies had different designs, we found that the easiest way to summarize all doses in the following categories (of note, most of the studies had a dose expansion protocol):

- For studies with **intravenous (IV) administration**, the dosage was given in one or more of the following categories.
    - **Cumulative dose range (Mbq/kg)** – which depicts the range of the cumulative dose of radiation received among the patients in the study group. Note that one patient has only one value of the "Cumulative Dose (Mbq/Kg)". Note also that the cumulative dose is given in "Mbq/kg", which makes it depended of the bodyweight of the patient (unlike the absolute cumulative dose)
    - **Dose range for one cycle (MBq/kg)** – which shows the radiation dose range for one cycle. All doses administered in one fraction/cycle in the study are in the depicted range.
    - **Absolute cumulative dose range (MBq)** – which depicts the range of which the absolute cumulative dose of radiation received among the patients in the study group. Note that one patient has only one value of the "Absolute Cumulative Dose (MBq)". Note also that the absolute cumulative dose is given in "MBq", which makes it not depended on the weight of the patient.

- For studies with **intralesional administration**, the dosage was given in one or more of the following categories.
    - **Absolute cumulative dose range (MBq)** – which depicts the range of which the absolute cumulative dose of radiation received among the patients in the study group.
    - **Dose range for one injection (MBq/Injection)** – which depicts the radiation dose range for one injection (intralesional administration).

In Table 3 note that not all the values in all categories are filled in. A white box indicates that the value was not given by the article.

Note also the big difference between doses given in the different studies.

5.5 Safety and efficacy results.

All the 13 articles described safety and efficacy data. Some in more detail than others. Systemizing the safety was a bit challenging for the following reasons:

(a) the patients across the different studies were being treated for different types of cancer, and that gives:

(a.i) different burdens of disease and symptoms,

(a.ii) different size and locations of the targeted loci for TAT-products,

(a.iii) different patient demographics.

(b) the administration method (intravenous / IV, intralesional) differs between studies;

(c) doses of radiation administered across patients varies:

(c.i) between patients in the same study,

(c.ii) between patient in different studies.

Regardless we have decided to summarize safety data as per the following.

The Safety data are summarized in TEAE (Treatment-Emerged Adverse Events) reported, as per CTCAE (Common Terminology Criteria for Adverse Events)(35), where AEs (Adverse Events) are classified into five severity grades (1 = mild, 2 = moderate, 3 = severe, 4 = life threatening, and 5 = death).

In the safety column, information is summarized through the following formulas:

- TEAE reported: - where the TEAEs given the different articles are listed, together with their respective severity grade.
- *() – where inside the brackets is written additional information about the study design considered important for the understanding of the safety data.
- (“”) – where inside is the authors (the author of the original article) interpretation of TEAEs.

The Efficacy data are summarized in objective responses as by RECSIT 1.1 (36). Note that the objective responses are described in different formats (e.g. CR, ORR, PFS…). Given that there were different types of cancers treated in across the studies, there is some variability in markers describing efficacy results. We choose to use the terminology of the original article to describe the efficacy data, to prevent distortion of the data. Abbreviations used on the efficacy column are expanded in the bottom row of Table 4.

The safety and efficacy data of the 13 articles summarized are summarized in Table 4.

Given the heterogeneity (of the patient groups; TAT used; dose range; and safety and efficacy data formats) across the 13 articles included, we chose to add one more Table (Table 5) in the data review. Table 5 summarizes the main conclusions from the original articles.

6.0 Discussion

6.1 Alpha-emitters and the consequences of their half-lives.

The alpha-emitters used in the 13 studies included are ^{212}Pb, ^{213}Bi and ^{225}Ac, with a respective half-life of approximately: 46 min (^{213}Bi), 10.6 hours (^{212}Pb) and 10 days (^{225}Ac).

The relatively short half-life of several alpha-emitters, (for example that of 46 min for ^{213}Bi) has given rise to some technical difficulties by limiting its use in many locations where the production cannot happen in house. This issue, together with the relatively high cost and low supply of alpha-emitting radionucleotides, has obstructed the development of TAT as a practice. Furthermore, even when production near the clinic is possible, and one manages to synthesize the alpha-vector-conjugates in time, this still leaves limited time for the TAT-compound to reach its designated target after administration in some cases. This being more of a problem for nucleotides with shorter half-lives, such as ^{212}Pb or ^{213}Bi.

To illustrate this issue we can look at reports by Cordier et al. (27). In their study, one patient had an astrocytoma (5 cm in diameter) and received ^{213}Bi-DOTA-substance P, locally through an intratumoral catheter. Substance P is a physiological ligand of NK-1 receptors, consistently overexpressed in gliomas, thus acting as a fine targeting vector. It was reported that: "*Radiographic changes after the injection, suggestive for radionecrosis, were observed mainly in proximity to the catheters in cortical and apical parts of the tumor, and only to a much lesser extent in deep and basal parts of the tumor*" (Figure 5, from (27)). The authors discuss this further and state that "*Small tumors tend to exhibit a complete radionecrotic appearance, whereas larger tumors seemed to be mainly necrotic in the proximity of the implanted catheters*."; and concludes with: "*Due to the relatively short half-life of ^{213}Bi, this concept probably has most of its therapeutic potential in the treatment of small, critically located gliomas*", as opposed to bigger tumors (27).

On the other hand, the short half-life of the radionucleotide combined with the short range of alpha radiation, should theoretically lead to less toxicity to undesignated areas and organs. This is also illustrated in the same study where the authors discuss that: " *SPECT/CT and blood sampling confirmed high retention of the radiopharmaceutical at the tumor site, in agreement with absence of toxicity to nontargeted, healthy brain tissue or other organs*."(27)

When designing a TAT-compound, the half-life of the chosen alpha-emitting nucleotide is of importance, as shown above. Not only in regard to the technical availability and price of the compound, but also as it alters the pharmacokinetics of the TAT-product.

6.2 Vectors and their importance

We have already discussed the importance of vectors in TAT in Chapter 3.3.5 of the Introduction, this chapter continue and elaborate this discussion.

In reference to the 13 articles in this review, we can see in Table 2 that both mAbs and ligands have been used as vectors. When cross referencing the vector used to the type of tumor the patients had on each respective study, we see that the vector chosen is often the same among different studies with same patient groups, for all studies except the mCRPC studies that use both ligands and mAbs. The vector selection is specifically made to alter and adjust the kinetics of the TAT-product to the desired target.

On the two leukemia studies (30,31), the vector used is a mAb. As mAbs are relatively large proteins, they have longer circulation time. This proves advantageous given that the targets of the TAT-product in leukemia are the bone marrow and the circulatory system. Monoclonal antibodies are thus perfect vectors to deliver the alpha-emitters (^{213}Bi and ^{225}Ac respectively, (30,31)) to their intended target (leukemic blasts). The vector used in both studies is Lintuzumab, an anti-CD33 monoclonal antibody. CD33 is a transmembrane receptor expressed on cells of myeloid lineage (37).

For the treatment of carcinoma in situ of the bladder with intravesical injections, the vector used is the study by Autenrieth et al., is also a mAb (33). Given that carcinoma in situ arises from, and is located in the epithelial tissue of the luminal side of the bladder, mAbs would also be advantageous. The TAT-product is delivered to the lumen of the bladder with its target is on the luminal side of the bladder wall. The radionuclide-mAb conjugate can reach its intended target as well as remain on the lumen of the bladder. As mAbs are large proteins their absorption in the circulatory system is limited, thus hindering side effects in other organ systems. This is supported by the safety reports, with no treatment emerged adverse events recorded among patients in this study (33).

For the glioma studies (27,29,32) the ligand used is Substance P. The physiological ligand of transmembraneous neurokinin type-1 receptors (NK-1) is substance P and WHO grades II–IV gliomas have been shown to consistently overexpress NK-1 receptors. Local intratumoural injection of radiolabeled substance P exploits this overexpression of the NK-1 receptor (27).

For the three studies on patients with mCRPC included in this review, both mAb (J951, mAb to PSMA) (34) and ligands (PSMA-617) are used (24,25). As discussed in chapter 3.3.5 the same differences in TEAEs reported, based on vector conjugated to beta-emitters also applies to alpha-emitters. Tagawa et al. (34) using mAb as a vector, reports lower occurrence of xerostomia (27%) while reporting one case on severe anemia. Kratochwil et al. , using a ligand as vector, reports that "*xerostomia is dose limiting before hematological toxicity becomes relevant*"(25). (Note that this also supports the argument and example presented in chapter 3.3.5)

For the included malignant melanoma studies the vector used is the same: 9.2.27 (23,26). The monoclonal antibody 9.2.27 (anti-NG2 or anti-neural/glial 2 antibody) is a perfect vector for targeting malignant melanoma because of its specificity for the human melanoma chondroitin sulfate proteoglycan (MCSP, also known as NG2), an antigen expressed by most melanomas (38)

Lastly, we have the NETs studies (6,28) using a ligand (the SSTR-targeting peptide - TATE) as vector. Most NETs strongly express somatostatin receptors (SSTR), providing the basis therapy with somatostatin analogs, one of which is TATE.

6.3 Administration

Choosing the right alpha-emitter and the right vector (and chelator) are not the only aspects that affects the safety and efficacy of a specific TAT-product. The administration method of the compounds is also of importance. The vision behind TAT is that it will specifically target tumors by itself. But if the patient were to have just one or few (not diffuse) tumors, one could theoretically help the targeting process and increase the efficacy of TAT, by delivering the product on site, through an intratumoral injection. Thus, increasing further the specificity of the delivered radiation dose, and potentially reducing side effects and increasing efficacy.

This is of course not always possible. With leukemia for example, we do not necessarily have a solid tumor. Furthermore, with disseminated cancer (and late-stage cancer) it would not be possible to inject numerous metastases one by one. Another aspect to think of is that intratumoral injections would also not take into account the treatment of micrometastases.

That is precisely the advantage of IV-administrated TAT-products. The TAT-products´ properties should enable it to find and fight off all cancer cells in the body, regardless how small or many they are.

But on the cases where the tumor is in fact solid, and we do have an overview of where the tumor is, an intratumoral administration may be beneficial.

In Table 2 (and all other tables, orange rows) we see that 5 of the 13 studies have chosen an intratumoral approach. When looking at safety reports, the intratumoral approach appears to be much safer, with 4 of the 5 studies reporting no related treatment emerged adverse effects (TEAEs) (23,27,32,33), and only reporting temporary TEAEs (29); in comparison to the 6 of the 7 studies with intravenous (IV) approach reporting different TEAEs. This supports the hypothesis that direct intratumoral injection should reduce side effects and toxicity.

6.4 Dosages of radiation, difference between articles

Table 3 summarize the doses of radiation delivered to patients among the 13 studies included in the current review. Most of these studies were on early stages of clinical trials (phase 1 studies, pilot studies or retrospective analysis of case series) and used dose expansion protocols. This means that across the studies but also in each study, not all patients received the same dose of radiation with their TAT injections.

Comparing doses between studies would be inefficient, as most of the studies have different designs (the TAT compound, its vector, the administration method), different patient demographics, as well as different types of tumors and burdens of disease. Table 3

serves as a summary of the doses to the reader's information. Doses of radiation will not be discussed further in this thesis.

6.5 Safety and efficacy.

6.5.1 General Limitations

The following aspects are considered as general limitations on the discussion of safety and efficacy of the articles included in this discussion.

(a) All thirteen studies are single-center studies that consist of small patient cohorts, ranged from five to 28 patients per study.
(b) Six of the 13 discussed studies are only available as abstracts. Despite that they are included in this discussion as per the reasons presented in Chapter 5.1.

6.5.2 Limitations on safety data review

When reviewing the safety data from the thirteen studies included in this discussion several considerations should be kept in mind:

(a) Even though all the studies explore the use of TAT, there are many variables that differ from study to study. Variables such as patient groups, type of cancer the patients are treated for, type of alpha-emitters used, type of vectors used, type of chelators used, administration methods, radiation doses, study designs and study follow up. This heterogeneity among the data across articles poses a significant obstacle to conducting a comparative analysis between the articles. Nevertheless, despite the aforementioned limitation, these articles still provide a valuable perspective on the safety profile of TAT in general, and represent a crucial window into comprehending and predicting the safety of specific TAT-products based on the components used and their underlying physical- and chemical properties.

(b) Most of the patients enrolled in the thirteen studies, have a high burden of disease that is of late stage (stage IV), metastatic or inoperable cancers (note that there are also patients with stage II, III cancer). Furthermore, it should be noted that a number of the patients had also previously received (or received the same time as TAT), other treatments know to carry a high risk of TEAEs, such as chemotherapy or other radiotherapy. This complicates how the emerged AEs (adverse events) are to be classified and interpreted. It poses a challenge to determine whether the emerged AEs are caused by the TAT-product, other therapeutic interventions, or simply because of the normal progression of the disease. We have relied on the authors' interpretation and classification of the TEAEs despite the potential risk of introducing their bias into our data analysis. We recognize that the original authors had a better overview of the factors mentioned in this paragraph, due to their familiarity to the patients. Therefore, we think that by adopting their understanding of the AEs, we aim to increase the likelihood of arriving at a more accurate conclusion.

(c) Only seven of the 13 studies we have included are full articles. The remaining six are only abstracts (as mentioned in chapter 5.1). This means that the safety (and

efficacy) data reported on those six articles is a form of summary or conclusion and does not contain the full dataset regarding safety. However, given our strategy to accept the original author´s conclusions as per paragraph (b), this should not be a hindrance in our safety analysis.

6.5.3 Limitations on efficacy data review

There are at least two limitations one should keep in mind when reviewing the efficacy data.

(a) Most of the patients involved in the 13 studies we discuss have late-stage cancer. Some of these patients have also previously (or at the same time as being given TAT) received other treatments to fight off their disease, including surgery, chemotherapy, or other radiotherapy. This could pose an issue regarding the efficacy analysis. However, most of the original authors have taken this in consideration and either established a timeline or highlighted the difference and impact that TAT has had.

(b) The 13 studies included in our discussion were on early stages of clinical trials (phase 1 trial, pilot study, retrospective analysis of case series). As a direct result of study protocols, there is a greater emphasis on assessing the safety rather the efficacy the treatment. Although this does not limit the reliability of the safety analysis reported, it does restrict the depth of the efficacy analysis. Since the doses and administration protocols for each TAT-product are not yet fully fine-tuned, further studies in the field would offer promising opportunities to enhance the efficacy of TAT and further optimize their therapeutic potential.

6.5.4 Safety and efficacy profiles based on type of cancer treated.

Across the 13 studies included, there are a total of six types of neoplasias patients are treated for (Table 2).

(a) Patients treatment for carcinoma in situ (bladder): 1 article (33);
(b) Patients treatment for glia tumors: 3 articles (27,29,32);
(c) Patients treatment for malignant melanomas: 2 articles (23,26);
(d) Patients treatment for leukemia: 2 articles (30,31);
(e) Patients treatment for mCRPC: 3 articles (24,25,34);
(f) Patients treatment for NETs: 2 articles (6,28).

When analyzing safety and efficacy profiles reported among studies that treat the same type of cancer, the following tables should be used as an overview and to cross reference results.

Table 2 (TAT-products and types of cancer treated)
Table 4 (Safety and efficacy data)
Table 5 (Authors conclusions)

(a) To start with, we have the article where treatment was administered to patients with Carcinoma in Situ of the bladder(33). According to rapports given, there were

no treatment emerged adverse effects (TEAEs). Given the cautious administration method, the fact that no TEAEs were reported was not unexpected. The TAT-product (^{213}Bi-cetuximab) was administered as an intravesical injection that was drained 120 min post injection. Of the 13 patients enrolled in this study, two patients had a complete remission, one had a complete remission with a relapse 15 months afterwards, and nine had no significant effect from the treatment. While not all patients benefited from the treatment, due to the lack of side effects the original author concludes with: *"Treatment of CIS of the bladder with the alpha-emitter 213Bi coupled to a mAb targeting EGFR was shown to be a safe treatment option without any adverse effects*"(33).

(b) The three articles where treatment was administering to patients with Glia Tumors (27,29,32) also report minimal TEAEs. Two of the articles report no TEAEs (27,32) and one article reports grade 2-3 temporary TEAEs (edema, epileptic seizures, aphasia)(29). All the three studies used an intratumoral administration method for TAT, with either ^{213}Bi or ^{225}Ac conjugated to Substance P. The study with the ^{225}Ac-Substance P conjugate reported the transient TEAEs (29). This could be due to the relative longer half-life of ^{225}Ac (10 days) as opposed to ^{213}Bi (46 min). However, all authors conclude with that the treatment is safe and relatively well tolerated.

When it comes to efficacy, all three articles report increased overall survival among their patients. Only one of the three studies (by Cordier et al. (27)) is a full article, and not only an abstract. Cordier et al. reports that *"Targeted therapy of critically located WHO grade II–IV gliomas with locally injected ^{213}Bi-DOTA-substance P is feasible and without relevant toxicity. Compared to therapeutic approaches using beta-emitters, the treatment of gliomas using short-range alpha-emitters may allow similar efficacy to be achieved with lower toxicity to healthy brain areas. Due to the relatively short half-life of ^{213}Bi, this innovative concept probably has most of its therapeutic potential in the treatment of small, critically located gliomas*" (27). The contrast in efficacy between the treatment of smaller and larger tumors can be clearly observed in Figure 5 and Figure 7, respectively. In Figure 7 (treatment of a smaller tumor) radionecrosis can be seen on all parts of the tumor, whereas in Figure 5 when treating a larger tumor radionecrosis is much more prominent in proximity to the injecting catheter.

Across the three articles treating Glia tumors, treatment seems to be effective with increased OS (overall survival) on patients, and radiographic proof of tumor necrosis.

(c) Two studies administering treatment to patients with malignant melanomas were included (23,26). Both use the same type of TAT (^{213}Bi - cDTPA - 9.2.27; with 9.2.27 being a mAb targeting melanoma chondroitin sulfate proteoglycan (anti-MCSP also known as anti-NG2). One study delivers the TAT intratumorally (23) and one via IV administration (26). Both studies reporting no toxicity and only minor TEAEs such as nausea and pain in the injection site. When discussing efficacy, it is easier to analyze each study separately.

Allen et al. (23) used intratumoral/intralesional administration on malignant melanomas and had all lesions excised 4 weeks post treatment to evaluate their histopathology. The following was reported: *"the results showed that the intralesional TAT is non-toxic and locally efficacious up to 1350 µCi with …almost complete cancer cell kill (at 16.65 MBq and above) with few viable cell clusters*". This proving the concept of radiotoxicity for locally injected TAT.

Raja et al. (26), used an IV approach as it was treating metastatic malignant melanomas. Generally, there was reported multiple tumor regressions, both for micro- and macro-metastases, but most patients relapsed before the 12th week post treatment, even though 13 of the 22 patients had a partial response to the treatment the first few weeks (Table 4 and (26)). By week 12, two patients had almost full remission while the others had relapsed. However, what was remarkable was the clear effect this treatment had on some of the patients. Histology works shows impressive regression of metastatic lung nodules at 8 weeks post TAT treatment in some of the patients. The author´s conclusion was *"No evidence of renal damage was observed up to 592 MBq over 12 months. TAT is therefore safe up to 592 MBq and in some patients an effective therapeutic approach for metastatic melanoma*." (26)

(d) The two studies with leukemia patients (30,31), both led by J. Jurcic, show generally a higher number of TEAEs, with a significant proportion of the reported TEAEs being more severe (grade 3, 4), such as neutropenia, neutropenic fever and thrombocytopenia. The hematological AEs can be explained due to the fact that the alpha-emitters used in these studies were coupled to anti-CD33 antibodies, with CD33 being an antigen expressed on cells of myeloid lineage. It is reasonable to expect at a reduced immune system when targeting myeloid cells, therefore the grade 4 TEAEs reported in these studies should be considered in that context. Of note, patients in the Jurcic et al. study receive TAT together with chemotherapy (low-dose cytarabine (LDAC)) (30). Note also that the abnormal white cell counts normalize after 2-4 weeks post treatment. Other TEAEs reported on the two leukemia studies include: infection, hypokalemia, fatigue and increased liver parameters, most of them being grade 1-2 TEAEs. Overall, the TEAEs was reported as manageable, and the TAT treatment is regarded as safe by the original authors.

Jurcic et al. reports: *"Although 213Bi-HuM195 killed large leukemic volumes in many patients, none achieved complete remission. Because of the nature of alpha particle radiation, complete remission at 30 days after treatment would have required the individual targeting and killing of 99.9% of the leukemia cells…. treatment with ^{213}Bi-HuM195 as a single agent would require extraordinarily high injected activities. On the other hand, because of the short range and the high linear energy transfer, alpha particle immunotherapy is ideally suited to the treatment of residual disease"* (31).

(e) Treatment to patients with mCRPC (metastatic castration-resistant prostate cancer) seems also to be well tolerated (24,25,34). Beside one patient having grade 4 anemia (considered unrelated to TAT, given that the patient had previously received 4 cycles of 177Lu-PSMA, a beta-emitter (34)), most of the reported TEAEs were grade 1 and

2. Reported TEAEs were xerostomia, pain, fatigue, nausea; with xerostomia being the most frequent issue. Kratochwil et al. (25) reports *"Severe xerostomia was dose-limiting before hematological toxicity became relevant".* Overall were the TEAEs reported as mild and manageable.

There was an objective response of PSA-decline in the range of 60-75% among the patients in each study. Kratochwil, who was first author in two of the mCRPC articles (24,25), goes as far as concluding with: *"A standard treatment dose of 100kBq/kgBW Ac-225-PSMA-617 administered every two month seems both associated with remarkable anti-tumor activity and tolerability. In our hospital it became the standard treatment activity for the first phase of routinely clinical application as a salvage therapy for mCRPC."*(25); and *"In regard to mCRPC alpha-emitters seem superior in comparison to beta-nuclides when tagged to the identical carrier molecule*"(24).

(f) Lastly there are two studies of patients with NETs (neuroendocrine tumors) (6,28). Both studies with Delpassand as first author. Both studies use the same TAT (212 Pb – DOTAMTATE, which utilizes a peptide ligand as a targeting molecule), delivered through IV administration. Both articles (6,28) report a number of TEAEs (170 and 144 respectively for each article), with most of the TEAEs being mild grade 1-2 (95% and 97% respectively for each article) and few being severe grade 3-4 (3% or 5% respectively for each article). The severe TEAEs (including renal failure; worsening achalasia; an acute cerebrovascular accident, hypoglycemia, asthma exacerbation, and septic shock leading to death) were not considered related to the treatment. Both articles by Delpassand et al. concludes that ^{212}Pb-DOTAMTATE appears to be well tolerated, with mild and manageable toxicity. Kidney toxicity being is the main issue that needs further investigation.

These studies have very promising results regarding efficacy. Not all patients showed a response, but most patients achieved stable disease (SD as per RECIST 1.1 is regarded as a response in patients with late-stage cancer/disseminated cancer as they otherwise would progress). Of the patients 12/20 and 7/10 respectively for (6) and (28) showed SD. Seven of 20 and two of 10 respectively had a partial response (PR as per RECIST 1.1); and one of 20 and one of 10 respectively had a complete remission. The patients that received higher doses showed generally a better response to treatment.

The PET scans of some of the patients are particularly exceptional. Figure 3 shows the ^{68}Ga-DOTATATE PET/CT scans from the MAD4 cohort that received the highest dose of ^{212}PbDOTAMTATE with 4 doses of 2.50 MBq/kg/cycle. A partial response with a median decrease in the sum of the diameters of the target lesions (SOD) of 41% was observed in all patients (6). Figure 4 depicts patient MAD4-02 that had the best response to treatment in the cohort at 85% decrease in SOD. Delpassand et al. concludes with *"^{212}Pb-DOTAMTATE is safe. Preliminary efficacy results are highly promising. If these results are confirmed in a larger, randomized, multicenter clinical trial, would provide a substantial benefit over currently FDA approved therapies for*

patients with metastatic or inoperable SSTR-expressing NETs regardless of the grade and location of the primary tumor" (6).

6.5.5 General safety profile analysis

The safety data from our 13 articles are summarized on **Table 4**, in the form of TEAEs reported, as per CTCAE (Common Terminology Criteria for Adverse Events)(35), where AEs (Adverse Events) are classified into five severity grades (1 = mild, 2 = moderate, 3 = severe, 4 = life threatening, and 5 = death). The reported adverse events summarized on Table 4, are the ones the original authors considered related to TAT treatment.

Five of the studies reported no TEAEs (23,24,27,32,33). Whereas the other studies report a number of TEAEs, most of them being mild (Grade 1-2) (Table 4).

The two studies performed in leukemia patients (30,31) have the most severe TEAEs. They show generally a higher number of TEAEs, with a significant proportion of the reported TEAEs being severe (Grade 4), such as neutropenia, neutropenic fever and thrombocytopenia. Nevertheless, that was to be expected as the alpha-emitters in these studies were coupled to anti-CD33 antibodies, that are expressed on cells of myeloid lineage. It is reasonable to expect at a reduced immune system when treating leukemia and targeting myeloid cells, therefore the Grade 4 TEAEs reported in these studies should be considered in that context. Of note, in one of the leukemia studies (30), patients receive TAT together with chemotherapy (cytarabine). Note also that the abnormal white cell counts normalize themselves after 2-4 weeks post treatment cycles.
Other TEAEs reported on the two leukemia studies include: infection, hypokalemia, fatigue and increased liver parameters, most of them being Grade 1-2 TEAEs.

For the remaining studies (6,25,26,28,28,29,34) there are mostly reported grade 1-2 TEAEs. Only other two studies (28,34) report grade 4 AEs (anemia (1 patient); septic shock and death (1 patient)), none of which were considered related to the TAT treatment. Both patients with the aforementioned severe AEs had previously received multiple cycles of beta-emitter based radiotherapy.

Of all the TEAEs, the most frequent ones are nausea and fatigue, followed by alopecia and xerostomia (on patients with mCRPC). However even the most common TEAEs (nausea and fatigue) have variable occurrence. For example while one study report 100% fatigue and 100% alopecia among its patients (28), others do not report these TEAEs at all.

Another interesting trend is that four of the five studies using a intratumoral administration approach (all Tables, orange rows) report no TEAEs, with only one article (29) reporting "mild temporary AEs (edema, epileptic seizures, aphasia)». Note that all patients in this report had previously received brain surgery and chemotherapy. It seems that a more direct administration method for the TAT (when possible) can be advantageous when it comes to safety profiles (as also discussed in Chapter 6.3).

All over, there were mostly mild and manageable TEAEs. The authors conclude that the TAT is most often well tolerated and the TEAEs are manageable (Table 5). Furthermore, the authors also state that TAT is a promising method but warrants further investigation.

6.5.6 General efficacy analysis

Considering the study designs of the 13 articles included (early stages of clinical trials), it is more interesting to discuss safety results then efficacy results. Nonetheless, we give a short summary on general efficacy trends in this chapter.

There is quite some variation in treatment outcomes among patients in the included studies. While some patients seem to have little to no effect from treatment, many patients seem to show a partial response, and some patients show remarkable results.

The fact that not all patients have the same benefit from the treatments is truly not discouraging as per for the following reasons:

(a) TAT is a form for personalized treatment, where not all patients are expected to qualify for treatment and benefit from it. Patients are dependent on their cancer (cells or stromal components) having expression of specific biologic targets, and furthermore have enough expression of these targets. The biological composition of cancer cells (and therein their specific biologic targets) between individuals is certainty heterogenous. As a result, some degree of variability in how well a vector targets these cells is expected, and with that comes variability in efficacy and treatment outcomes. TAT is definitely not a “one shoe fits all” type of treatment.

(b) Most studies included have dose-expansion protocols and are on early stages of clinical trials focusing on the safety aspects of the treatment. Treatment doses given to patients enlisted are not fully optimized yet. There is trend that patients with higher doses show better outcomes, as for example in (6), where MAD4 patients (receiving the highest dose among patients in the same study) had the best results. It would be logical to expect better efficacy with better dose regimes. Further phase 2 and 3 studies should provide with better treatment regimens and increase efficacy outcomes in patients.

(c) Among most pharmaceuticals and treatments in clinical medicine, including oncology, numbers need to treat are rarely one. It is not uncommon to have patients that do not respond to treatment. If some patients have effect from the treatment, that still gives enough indication that the drug is worth pursuing.

For the abovementioned reasons we choose to focus on the few exceptional results in this efficacy analysis, as they act as a proof-of-concept and provide evidence of the possibilities that TAT practice could lead to.

Some very interesting examples are the results of the MAD4 cohort patients from Delpassand study (6). Figure 3 illustrate how most patients with metastatic NETs show remarkable tumor regression, after four cycles with ^{212}Pb-DOTAMTATE. In the MAD4 cohort, the ORR by RECIST 1.1 was 80% and the median decrease in SODs for all patients was 41%

(sum of the diameters of the target lesions) (6). Figure 4 illustrates the impressive regression with 85% decrease is SOD in patient MAD4-02 after four cycles with ^{212}Pb-DOTAMTATE.

Another interesting result is from the study by Allen et al. (23) with intralesional injections in malignant melanomas. All lesions were later resected at 4 weeks post treatment, and their histopathology was evaluated. It was reported that histopathological investigation shows *"…almost complete cancer cell kill (at 16.65 MBq and above)…"*. This proving the concept of radiotoxicity in vivo for TAT, in this case via intralesional administration.

Generally, when viewing efficacy results (Table 4), it seems that TAT has some effect across all studies, with some variation. Patients receiving higher doses, tend to have the most effect. TAT does not seem to be a miraculous treatment granting complete remission for all patients. However, there is clear indication of cancer-regression in the majority of patients. This fact should be enough indication for further investigation of different TAT approaches. When we also take in consideration the remarkable effect it has had a few patients (patients with CR), then we most definitely should have enough evidence that the concept behind TAT works, and that it would be beneficial to further investigate and optimize this practice.

7.0 Conclusion

Targeted Alpha-emitter Therapy (TAT) is not one single product, but a method under the umbrella term of radiotherapy. While the theory behind and similarities among product-components may provide some degree of predictability in properties and outcome, it is crucial to comprehend and thoroughly test each TAT-product independently.

That said, as far as we can go to conclusions from the few articles discussed in the thesis, TAT-products seems to have little toxicity with manageable side effects, with some variety in the severity of TEAEs, most of which are mild (grade 1-2) (Table 4). Regarding efficacy there is also some variability. The majority of patients show partial response to treatment, some patients have a minor effect of treatment, while others show remarkable results (Table 4, Figure 3 and 4). As expected, due to the heterogeneity of patient populations, TAT compounds and study designs across studies, efficacy rapports differ moderately. Still all studies tend to show promising results.

Most studies had a dose expansion protocol. There was a trend observed, where the patients receiving higher doses of radiation, had the better treatment results. This could indicate that that further inquiry into optimizing patient selection and dosage protocols should improve efficacy outcomes. Future prospective studies are needed to investigate this hypothesis.

Particularly interesting is the fact that all authors from the original articles included in this literature review seem to report that TAT is relatively well tolerated and shows promising results.

Kratochwil et al. goes as far as saying that *“Ac-225-PSMA-617 (a targeted alpha-emitter) seems to be both associated with remarkable anti-tumor activity and tolerability. In our hospital it became the standard treatment activity for the first phase of routinely clinical application as a salvage therapy for mCRPC.”(25)*

Will TAT revolutionize treatment outcomes for disseminated cancer? It's difficult to say for sure, but the theory behind it, as well as early preliminary results suggest that it might have a chance to do that. I personally believe that there is more than enough evidence to inspire further investigation and research.

8.0 Figures and Tables

Figure 1: Six decay schemes (**a**–**f**), including most alpha-emitters relevant for radionuclide therapy. All branching ratios larger than 0.1% are included. The beta- and alpha-particle energies given are for the highest intensity emission.

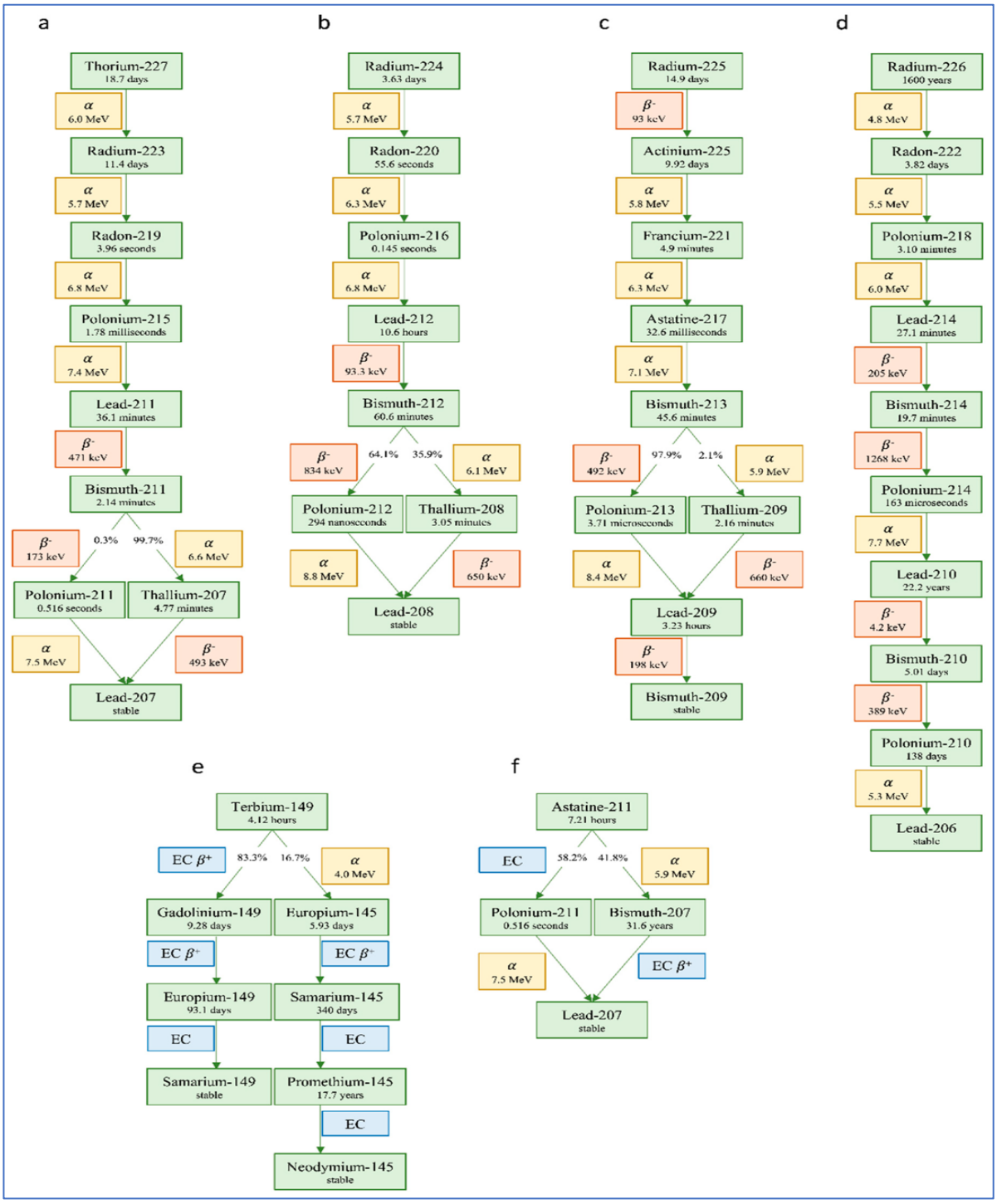


Figure 2: Article Inclusion strategy

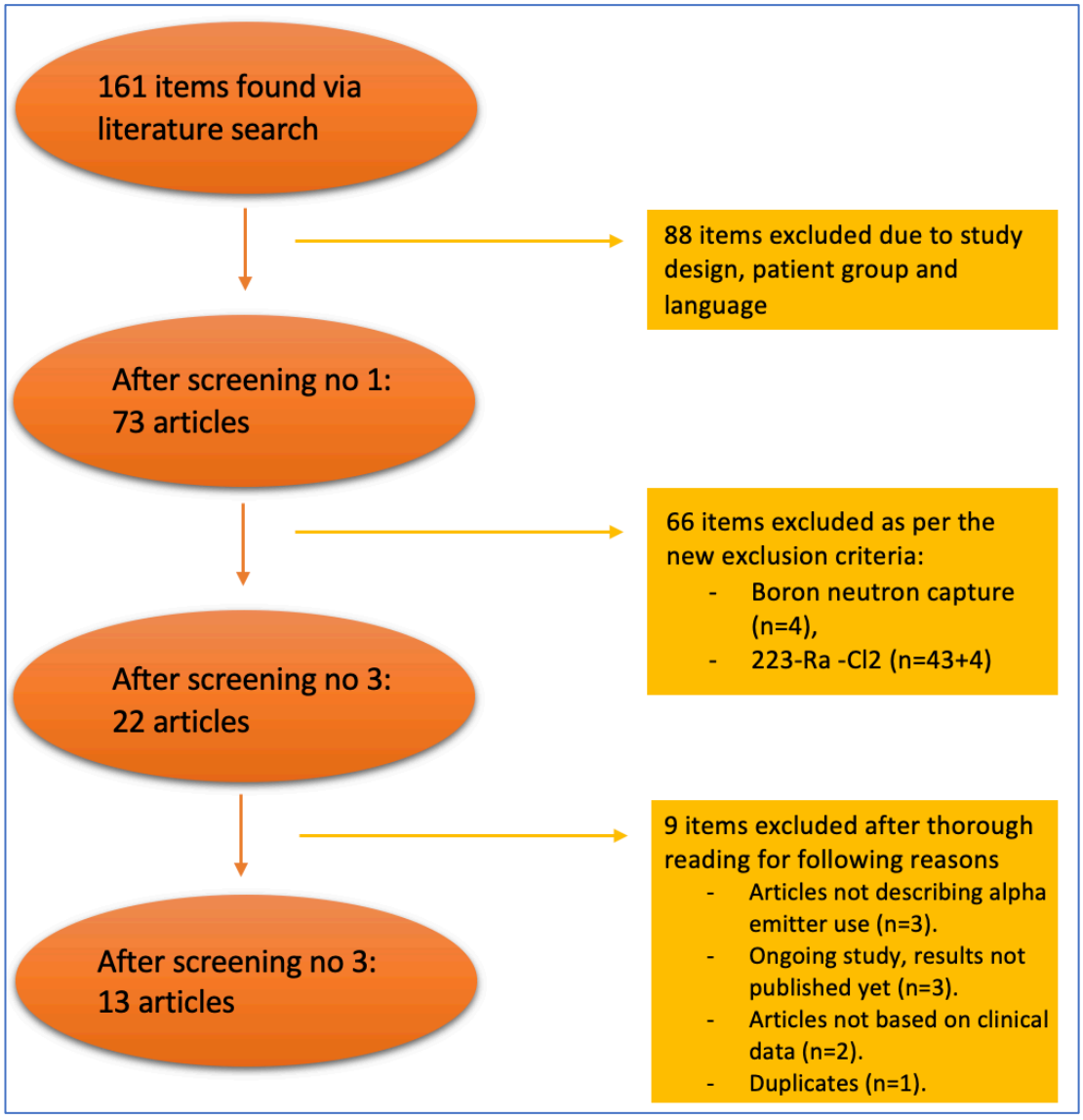


Figure 3: Volume rendered images of 68Ga-DOTATATE PET/CT scans from the first 10 subjects enrolled in cohort 4 (MAD4), before (image on the left) and after treatment (image on the right) with 4 cycles of 212PbDOTAMTATE at a dose of 2.50 MBq/kg (67.6 µCi/kg), for each cycle.

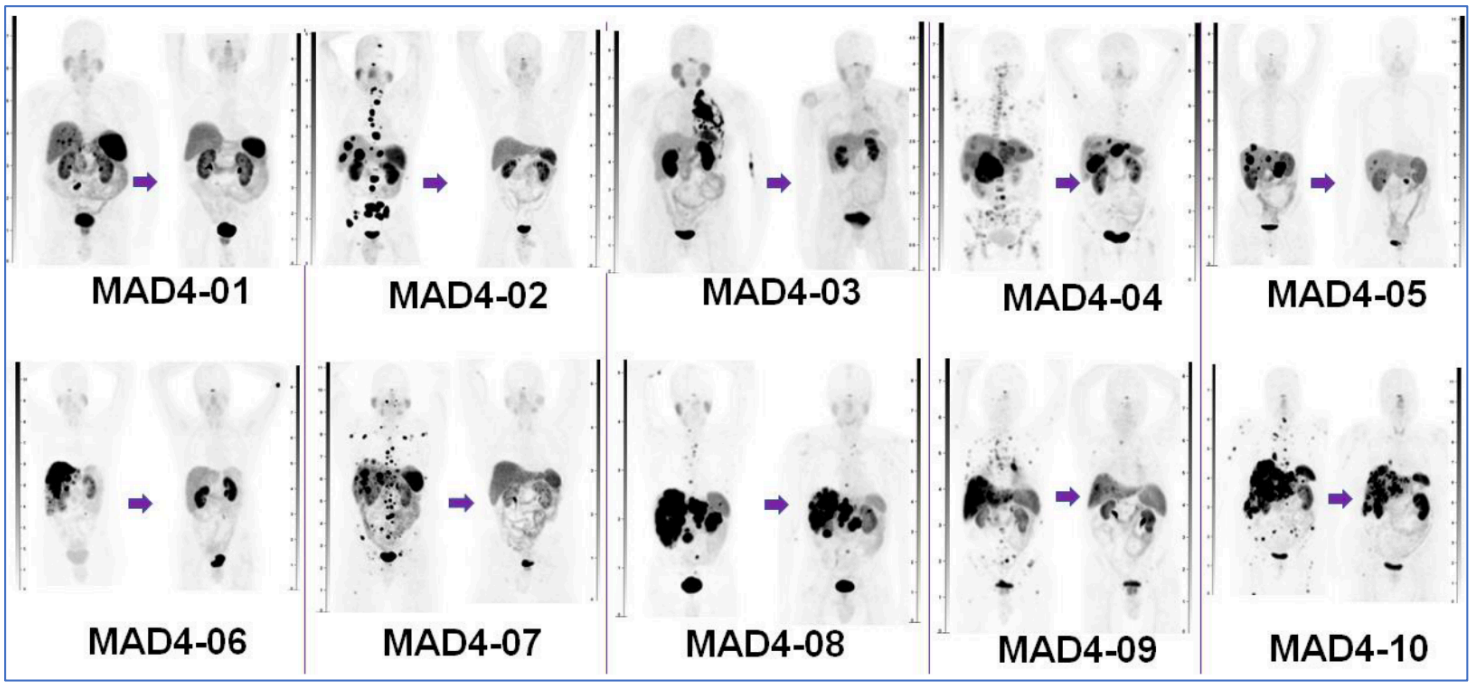


Figure from Delpassand et al. article (6), a free PMC article, Copyright © 2022 by the Society of Nuclear Medicine and Molecular Imaging.

Figure 4: Before and after images of 68Ga-DOTATATE PET/CT of MAD4-02 patient with metastatic bronchial carcinoid after treatment with four cycles of 212PbDOTAMTATE

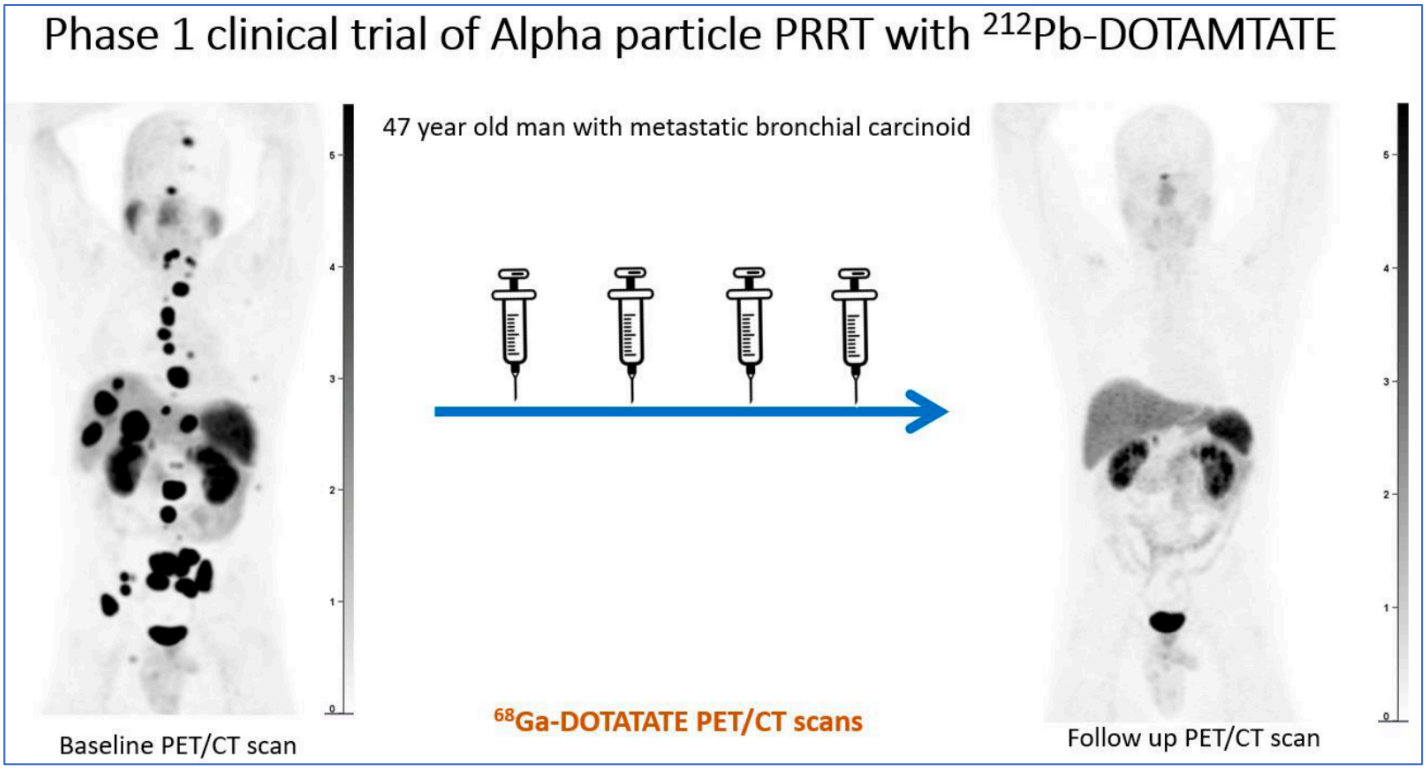


Figure from Delpassand et al. article (6), a free PMC article, Copyright © 2022 by the Society of Nuclear Medicine and Molecular Imaging.

Figure 5: MR images from patient 3, receiving intratumoral TAT injection, with subtext from original article.

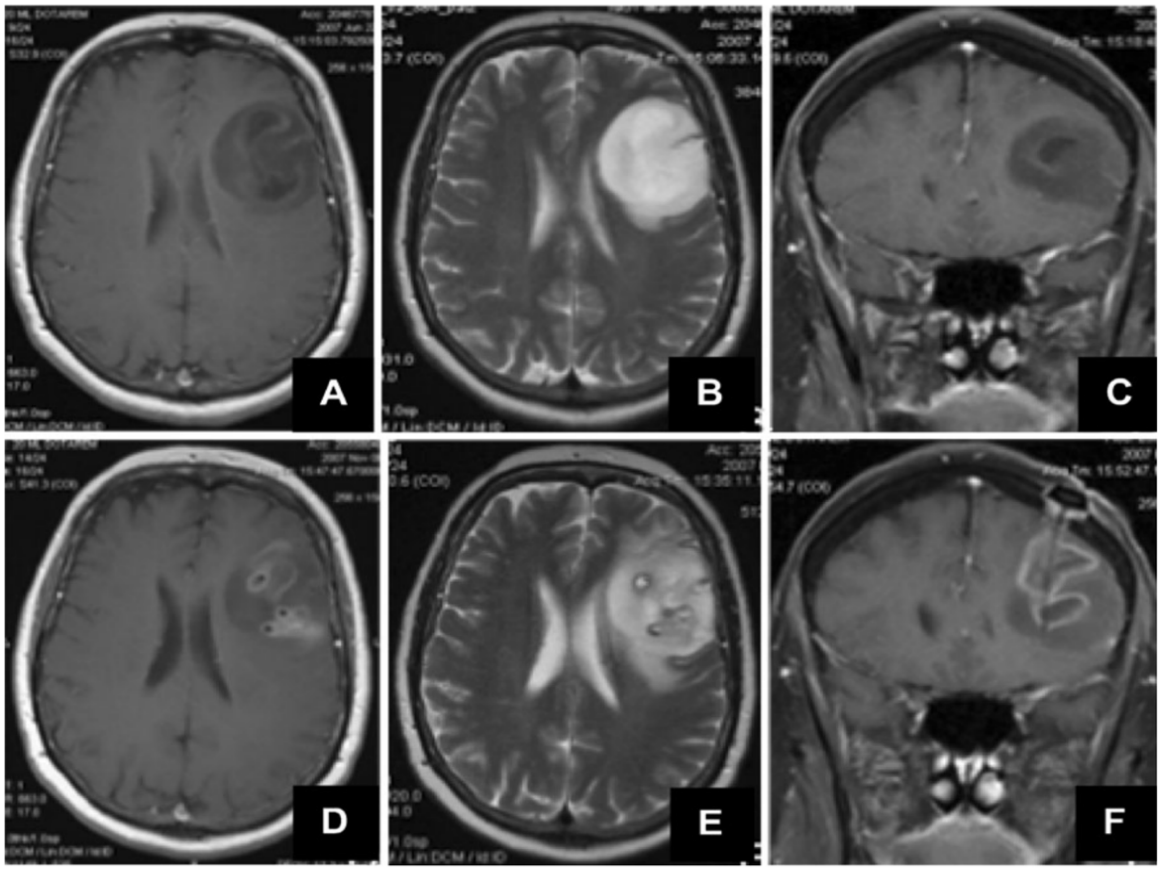


Fig. 5 Patient 3, left opercular anaplastic astrocytoma, contrast-enhanced T1- and T2-weighted MR imaging. **a–c** Initial imaging before stereotactic biopsy and catheter placement. **d–f** Status after radiopeptide treatment as described in the text. All three implanted catheter systems are visible in the axial planes; one catheter with its injection port is visible in the coronal plane. Note the inhomogeneous radionecrotic changes mainly concentrated around the catheters

Figure 7: MR images from patient 5, receiving intratumoral TAT injection, with subtext from original article.

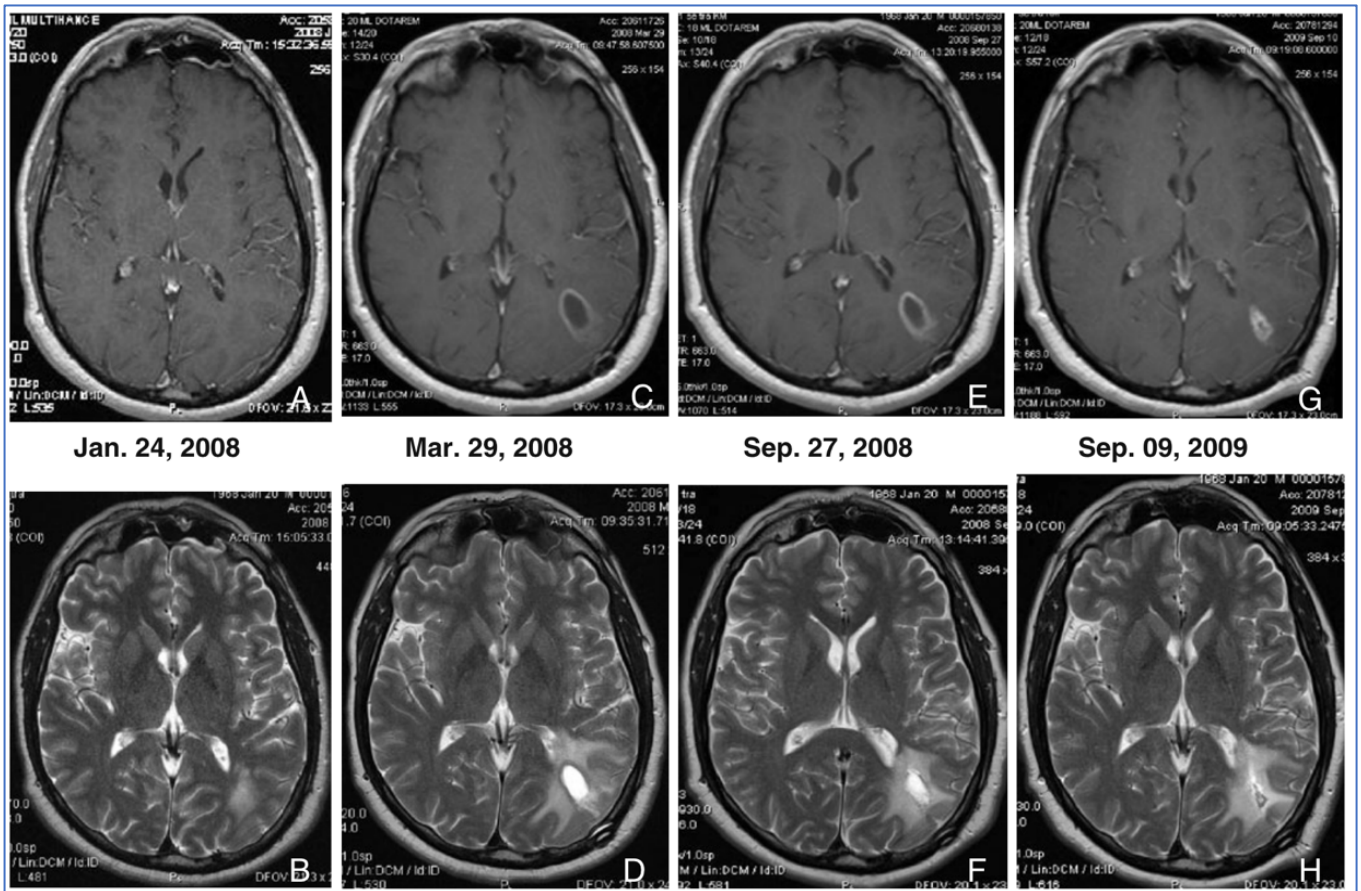


Fig. 7 Patient 5, left occipital WHO grade II astrocytoma, contrast-enhanced T1- and T2-weighted MR imaging. **a**, **b** Imaging before stereotactic biopsy and catheter placement. **c**, **d** Status 4 weeks after intratumoural ^{213}Bi-DOTA-substance P injection. **e**, **f** Status 7 months after radiopeptide injection; note that the lesion appears slightly more compact. **g**, **h** Imaging 19 months after radiopeptide treatment, most probably consistent with local scar tissue formation

Figure 6: Different types of radiation.

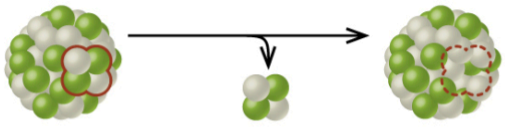
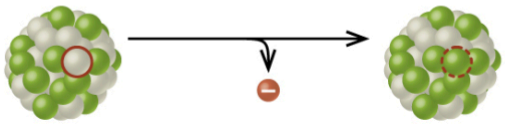
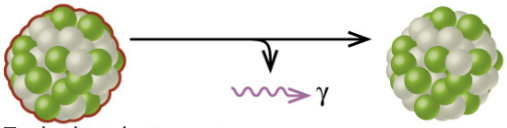
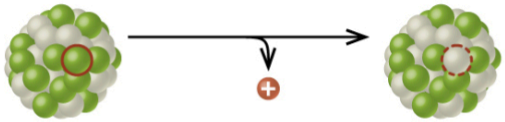
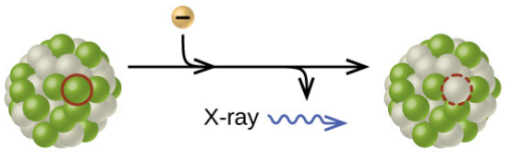
Type	Nuclear equation	Representation	Change in mass/atomic numbers
Alpha decay	${}^{A}_{Z}X \rightarrow {}^{4}_{2}He + {}^{A-4}_{Z-2}Y$		A: decrease by 4 Z: decrease by 2
Beta decay	${}^{A}_{Z}X \rightarrow {}^{0}_{-1}e + {}^{A}_{Z+1}Y$		A: unchanged Z: increase by 1
Gamma decay	${}^{A}_{Z}X \rightarrow {}^{0}_{0}\gamma + {}^{A}_{Z}Y$		A: unchanged Z: unchanged
Positron emission	${}^{A}_{Z}X \rightarrow {}^{0}_{+1}e + {}^{A}_{Y-1}Y$		A: unchanged Z: decrease by 1
Electron capture	${}^{A}_{Z}X \rightarrow {}^{0}_{-1}e + {}^{A}_{Y-1}Y$		A: unchanged Z: decrease by 1

Table 1

Article Referance Nr	Title	All authors	Type of study	Article size	Publication Medium	City/Country of study	Publication Year
(33)	Treatment of carcinoma in situ of the urinary bladder with an alpha-emitter immunoconjugate targeting the epidermal growth factor receptor: a pilot study	M.e, Autenrieth; C, Seidl; F, Bruchertseifer; T, Horn; F, Kurtz; B, Feuerecker; C, D'Alessandria; C, Pfob; S, Nekolla; C, Apostolidis; S, Mirzadeh; J.e, Gschwend; M, Schwaiger; K, Scheidhauer; A, Morgenstern	Pilot study	Full article	European Journal of Nuclear Medicine and Molecular Imaging	Munich, Germany	2018
(32)	Secondary glioblastoma multiforme-local alpha emiters targeted therapy with 213Bi-DOTA-substance P	L, Krolicki; A, Morgenstern; J, Kunikowska; H, Koziara; B, Krolicki; M, Jakucinski; D, Pawlak; C, Apostolidis; F, Bruchertseifer	Retrospective analysis of case series (dose escalation experience)	Only abstract	European Journal of Nuclear Medicine and Molecular Imaging	Warszawa, Poland	2016
(29)	Targeted alpha therapy of recurrent glia tumors: Clinical experience with Ac-Substance-P	F, Bruchertseifer; L, Krolicki; J, Kunikowska; H, Koziara; B, Krolicki; M, Jakucinski; D, Pawlak; A, Apostolidis; A, Morgenstern	Retrospective analysis of case series	Only abstract	NuklearMedizin	Warszava, Poland	2020
(27)	Targeted alpha-radionuclide therapy of functionally critically located gliomas with 213Bi-DOTA-[Thi8,Met(O2)11]-substance P: a pilot trial	D, Cordier ; F, Forrer; F, Bruchertseifer; A, Morgenstern; C, Apostolidis; S, Good; J, Muller-Brand; H, Macke; J.C, Reubi; A, Merlo	Pilot study	Full article	European Journal of Nuclear Medicine & Molecular Imaging	Basel, Switzerland	2010
(23)	Intralesional targeted alpha therapy for metastatic melanoma	B.J, Allen; C, Raja; S, Rizvi; Y, Li; W, Tsui; P, Graham; J, Thompson; R, Reisfeld; J, Kearsley; A, Morgenstern; C, Apostolidis	Phase 1 trial (open label, not randomized, dose escalation)	Full article	Cancer Biology & Therapy	Sydney, Australia	2005
(26)	Interim analysis of toxicity and response in phase 1 trial of systemic targeted alpha therapy for metastatic melanoma	C, Raja; P, Graham; S.m, Abbas Rizvi; E, Song; H, Goldsmith; J, Thompson; A, Bosserhoff; A, Morgenstern; C, Apostolidis; J, Kearsley; R, Reisfeld; B.j, Allen	Phase 1 trial (open label, not randomized, dose escalation)	Full article	Cancer Biology and Therapy	Australia (multicenter)	2007
(30)	Phase I trial of alpha-particle immunotherapy with 225Ac-lintuzumab and low-dose cytarabine in patients age 60 or older with untreated acute myeloid leukemia	J, Jurcic; M, Levy; J, Park; F, Ravandi; A, Perl; J, Pagel; B, Smith; J, Orozco; E, Estey; H, Kantarjian; D, Cicic; D, Scheinberg	Phase 1 trial (open label, not randomized, dose escalation)	Full article	Journal of Nuclear Medicine	Philadelphia, USA	2017
(31)	Targeted alpha particle immunotherapy for myeloid leukemia	J.g, Jurcic; S.m, Larson; G, Sgouros; M.r, McDevitt; R.d, Finn; C.r, Divgi; A.m, Ballangrud; K.a, Hamacher; D, Ma; J.l, Humm; M.w, Brechbiel; R, Molinet; D.a, Scheinberg	Phase 1 trial (open label, not randomized, dose escalation)	Full article	Blood	New York, USA	2002
(34)	Dose-escalation results of a phase i study of 225Ac-J591 for progressive metastatic castration resistant prostate cancer (mCRPC)	S.t, Tagawa; J, Osborne; M.j, Niaz; S, Vallabhajosula; P.j, Vlachostergios; C, Thomas; A.m, Molina; C.n, Sternberg; S, Singh; E, Fernandez; J, Babich; D.m, Nanus; K.v, Ballman; N.h, Bander	Phase 1 trial (open label, not randomized, dose escalation)	Only abstract	Journal of Clinical Oncology	New York, USA	2020
(24)	Ac-225-PSMA-617 for PSMA targeting alpha-radiation therapy of 28 patients with mCRPC	C, Kratochwil; F, Bruchertseifer; F.l, Giesel; C, Apostolidis; U, Haberkorn; A, Morgenstern	Retrospective analysis of case series	Only abstract	European Journal of Nuclear Medicine and Molecular Imaging	Heidelberg, Germany	2016
(25)	Dose escalation experience with Ac-225-PSMA-617 in PSMA targeting alpha-radiation therapy of patients with mCRPC	C, Kratochwil; F, Bruchertseifer; F.l, Giesel; C, Apostolidis; U, Haberkorn; A, Morgenstern	Retrospective analysis of case series (dose escalation experience)	Only abstract	European Journal of Nuclear Medicine and Molecular Imaging	Heidelberg, Germany	2016
(6)	Targeted alpha-Emitter Therapy with 212Pb-DOTAMTATE for the Treatment of Metastatic SSTR-Expressing Neuroendocrine Tumors: First-in-Humans Dose-Escalation Clinical Trial	E.s, Delpassand; I, Tworowska; R, Esfandiari; J, Torgue; J, Hurt; A, Shafie; R, Nunez	Phase 1 trial (open label, not randomized, dose escalation)	Full article	Journal of nuclear medicine : official publication	Hustion, Texas	2022
(28)	Targeted alpha-emitter therapy with 212Pb-DOTAMTATE in neuroendocrine tumor subjects who progressed following prior)OY-PRRT	E, Delpassand; R, Esfandiari; I, Tworowska; J, Torgue; J, Daniel Hurt; R, Nunez	Phase 1 trial (open label, not randomized)	Only abstract	Journal of Clinical Oncology	Huston, Texas	2022

Table 2

Article Referance Nr	No of patients (in final data review)	Alpha Emitter (+ Chelator) + Vector (mAb)	Alpha Emitter (+ Chelator) + Vector (ligand)	Intravenous	Type of Cancer (of patients enrolled)
(33)	12 patients	213Bi - p-SCN-Bn-CHX-A DTPA - cetuximab (mAb, anti-EGFR)		Intralesional (*Intravesical)	Carcinoma (Bladder; High Grade Carcinoma in Situ)
(32)	7 patients		213Bi - DOTA - substance P (ligand)	Intralesional	Glioma (GBM, Gliobalstoma multiforme, secondary)
(29)	13 patients		225Ac - Substance-P (ligand)	Intralesional	Glioma (WHO Grade II-IV)
(27)	5 patients		213Bi - DOTA - substance P (ligand)	Intralesional	Glioma (WHO grades II–IV)
(23)	16 patients	213Bi - cDTPA - 9.2.27 (mAb to melanoma chondroitin sulfate proteoglycan (MCSP))		Intralesional	Melanoma (stage IV)
(26)	22 patients	213Bi - cDTPA - 9.2.27 (mAb)		Intravenous	Melanoma (Metastatic, stage IV)
(30)	18 patients	225Ac - Lintuzumab (mAb, anti CD33)		Intravenous	Leukemia (Acute Myeloid)
(31)	18 patients	213Bi - SCN-CHX-A-DTPA - Litinzumab (mAb, anti CD33, HuM195)		Intravenous	Leukemia (AML, CMMOL, CML)
(34)	22 patients	225Ac - J591 (mAb to PSMA)		Intravenous	mCRPC (Castration-resistant prostate cancer with metastases)
(24)	28 patients		225Ac - PSMA-617 (ligand)	Intravenous	mCRPC (Castration-resistant prostate cancer with metastases)
(25)	10 patients		225Ac - PSMA-617 (ligand)	Intravenous	mCRPC (Castration-resistant prostate cancer with metastases)
(6)	20 patients		212Pb - DOTAM- TATE (ligand; TATE: SSTR targeting peptide)	Intravenous	NETs
(28)	11 patients		212Pb - DOTAM- TATE (ligand; TATE: SSTR targeting peptide)	Intravenous	NETs (pancreas (4), small bowel (3), midgut (1), ileum (1), thymus (1), and lung (1))

Table 3

Article Reference Nr	Safety (TEAE / treatment-emerged adverse events / considered by author related to TAT treatment)	Efficacy (Objective Responses to Treatment)
(33)	* (intravesical injection of TAT, drained after 120 min) No TEAE reported	Objective responses: 3/12 patients (2 patients CR; 1 patient CR + relapse 15 months after, 9 patients no effect)
(32)	No TEAE reported ("No any serious adverse reactions was shown, ...is safe and well tolerated")	Objective response: Overall Survival (OS) with TAT 46.8 months (median) (OS without treatment 7.8 months (median), OS with surgery and radiotherapy 27.1 months (median))
(29)	TEAE reported: Grade 2-3 temporary ("edema, epileptic seizures, aphasia")	Objective responses: PFS median 4 months (Group A (Gr II-III Gliomas) median PFS: 4.8 months; Group B (Gr IV Glioma) median PFS: 3 months)
(27)	No TEAE reported ("one patient temporary fatigue and transient facialis paresis, considered not related to TAT")	(Subjective response: Small tumors exhibited a complete radionecrotic appearance, whereas larger tumors seemed to be mainly necrotic in the proximity of the implanted catheters.)
(23)	No TEAE reported ("apart from pain during injection (intense but brief / lasting 3-4 s")	Objective responses: Histology almost complete cancer cell kill (at 16.65 MBq and above) with few viable cell clusters (all tumors were was excised 4 weeks post treatment)
(26)	No toxicity. TEAE reported: only nausea (Grade 1) for one patient. ("creatinine/GFR in normal range all patients")	Objective responses: Week 2: (13/22 patients, stable disease; 3/22 patients, partial response;) Week 4: (6 patients, partial response.) Week 8: (4 patients, partial response.) Weeks 12: (2 patients partial response, almost full remission). Multiple tumor regression (both micro and macrometastases, but sickness relapse after treatment for most patients)
(30)	* (together with other chemo (low dose cytarabine)) TEAE reported : Grade 4 (neutropenia (n=5 / 28%), thrombocytopenia (n=9 / 50%), febrile neutropenia (n=6 / 33%)), Grade 1-3 (pneumonia (n=5 / 28%), other infections (n=3 / 16%), atrial fibrillation/syncope (n=1 / 5%), transient creatinine increase (n=1 / 5%), generalized fatigue (n=1), hypokalemia (n=1), mucositis (n=1), rectal hemorrhage (n=1))	Objective responses: (2 CR, 1 CRp, 2 CRi) in 5/18 patients (28%), only at doses ≥ 0.037 Mbq/kg/fraction. (Response after 1 cycle (mean, TAT+Chemo) opposing to 3 (mean, chemo only).)
(31)	TEAE reported (transient): Grade 1 liver function abnormalities (22%, 4 patients), Grade 2 hyperbilirubinemia (11%, 2 patients), grade 3 leukopenia (11%, 2 patients) or 4 Grade 4 leukopenia (61%, 11 patients), neutopenic fever (66%, 8 patients). Median time for resolution of leukopenia 22 days (range 12-41 days)	Objective responses: (15/18 patients had leukemic blasts in the peripheral blood before treatment.) 14/15 (93%) had reduced circulating blasts (mean reduction 90%). Suppression lasted a median of 19 days (range, 8-42 days). 14/18 patients (78%) had reductions in the percentages of bone marrow leukemia cells
(34)	TEAE reported: Low Grade 1-2 temporary AE's (16 (73%) with fatigue, 11 (50%) pain, 11 (50%) nausea, 6 (27%) with xerostomia (5 of 6 with prior 177Lu-PSMA), 3 (14%) AST elevation.)	Objective responses: 60% (any PSA decline); 35% (>50% PSA decline)
(24)	No TEAE reported ("Hematological toxicity was tolerable")	Objective response: 20% of patients CR (after TAT); Week 8: 95% of patients PSA response (Ac-225); Week 24: 75% of patients PSA response (Ac-225). (vs 75% PSA response (Lu-177), weeks 8 50% (Lu-177))
(25)	TEAE reported: Xerostomia (Grade 2) ("Severe xerostomia was dose-limiting before hematological toxicity became relevant. 3 patients with 200kBq/kg and 1 patient with 150kBq/ kg requested dose reduction.")	Objective response: 70% of patients had PSA-decline of >50% (doses above 0.1 MBq/ kg/cycle)
(6)	TEAE reported (170) : Grade 1, Grade 2 (29%), Grade 3 (5%), Grade 4 (0%) Frequent TEAE: nausea (31%), fatigue, alopecia (25%)	Objective responses: (SD 12/20; PR 7/20; CR 1/20 as by RECIST1.1); patients with higher doses showing remarkable PET response. Median SOD (decrease of sum of the diameters was 41%.)
(28)	TEAE reported (144) : Grade 1 (79%), Grade 2 (16%), Grade 3/4 (3%) (3 serious TEAE (achalasia, asthma exacerbation, and septic shock, inkl 1 death (septic shock) / considered not related to study drug) Frequent TEAE :alopecia(100%), fatigue (100%), nausea (91%), anemia (36%), ALAT increased (36%), ASAT increased (36%) and lymphopenia (46%)	Objective responses: ORR 3/10 patient (1 CR, 2 PR, 7 SD as by RECIST1.1) s, PET response 7/10
	ORR (Overall Response Rate), PFS (Progression Free Survival), OS (Overall Survival), CR (Complete Remission) PR (Partial Remission) SD (Stable Disease), CRp (Complete Remission with incomplete platelet recovery), CRi (Complete Remission with incomplete count recovery)	

Table 4

Article Reference Nr	Alpha emitter type + vector	Cumulative Dose Range (between patients) (Mbq/kg)	Dose range for one cycle (MBq/kg)	Absolute Cumulative Dose Range (between patients) (Mbq)	Dose range for one injection (Mbq/Injection)	TAT administration method
(33)	213Bi - anti-EGFR	_	_	366 - 1 335	366-281	Intralesional
(32)	213Bi - substance P	_	_	4 000 - 16 000	2000	Intralesional
(29)	225Ac - Substance-P	_	_	20 - 280	20 - 40	Intralesional
(27)	213Bi - substance P	_	_	1 070 -7 360	1070 -2000	Intralesional
(23)	213Bi - 9.2.27	_	_	5.5 - 49.95	5.5 - 49.95	Intralesional
(26)	213Bi - 9.2.27	_	? - 9	55 - 1 184	_	Intravenous
(30)	225Ac - Lintuzumab	0.037 - 0.148	0.0185 - 0.074	_	_	Intravenous
(31)	213Bi - HuM195	_	10.36 - 37	602 - 3 515	_	Intravenous
(34)	225Ac - J591	0.0133 - 0.093	_	_	_	Intravenous
(24)	225Ac - PSMA-617	_	_	_	_	Intravenous
(25)	225Ac - PSMA-617	_	0.05 - 0.2	_	_	Intravenous
(6)	212Pb - DOTAMTATE	1.13 - 10	1.13 - 2.5	81-847 (mean 791)	_	Intravenous
(28)	212Pb - DOTAMTATE	2.5 - 10	2.5 - 2.5	(mean 773.3)	_	Intravenous

Table 5

Article Reference nr	Authors Conclusions
(33)	Treatment of CIS of the bladder with the alpha-emitter 213Bi coupled to a MAb targeting EGFR was shown to be a safe treatment option without any adverse effects.
(32)	Treatment of secondary GBM with 213Bi-SP is safe and well tolerated
(29)	Intracavitary/intratumoral injection of 225Ac-substance P is tolerated well. Only mild temporary adverse effects were observed. 225Ac use is promising and requires further clinical trials.
(27)	Targeted therapy of critically located WHO grade II–IV gliomas with locally injected 213Bi-DOTA-substance P is feasible and without relevant toxicity. Due to the relatively short half-life of 213Bi (46 min), this innovative concept probably has most of its therapeutic potential in the treatment of small, critically located gliomas
(23)	Intralesional TAT is non-toxic and locally efficacious up to 49950 MBq (1350 µCi)....histology showed almost complete cell kill at 16650 MBq (450 µCi) and above with few viable cell clusters.
(26)	No evidence of renal damage was observed up to 592 MBq over 12 months. TAT is therefore safe up to 592 MBq and in some patients an effective therapeutic approach for metastatic melanoma.
(30)	Fractionated-dose 225Ac-linutuzmab can be safely combined with LDAC and induce remission in older patients with untreated AML.
(31)	Absorbed dose ratios between the bone marrow, liver, spleen, and the whole body were approximately 1000 times higher for 213Bi-HuM195 than those for the B-emitting constructs."; "Although 213Bi-HuM195 killed large leukemic volumes in many patients, none achieved complete remission...Treatment of overt leukemia with 213Bi-HuM195 as a single agent would require extraordinarily high injected activities....a-particle immunotherapy is ideally suited to the treatment of residual disease.
(34)	PSMA-targeted alpha-emitter 225Ac utilizing intact Ab J591 is well tolerated with early evidence of clinical activity in a pre-treated population, including the majority with prior 177Lu-PSMA.
(24)	mCRPC alpha-emitters seem superior in comparison to beta-nuclides when tagged to the identical carrier molecule.
(25)	A standard treatment dose of 100kBq/kg administered every two month seems both associated with remarkable anti-tumor activity and tolerability.
(6)	Targeted alpha therapy with 212Pb-DOTAMTATE has been shown to be well-tolerated. Preliminary efficacy results are highly promising.
(28)	The use of 212Pb-DOTAMTATE in the recurrent setting is highly effective with manageable toxicity and warrants further investigation.

9.0 Bibliography

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